PHYSICO-CHEMICAL PROPERTIES OF CHICKPEA FLOUR, STARCH AND PROTEIN FRACTIONS AND THEIR UTILIZATION IN LOW-FAT PORK BOLOGNA

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By

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ABSTRACT

The main objective of this research was to investigate possible uses of Western-Canadian grown chickpea (*Cicer arietinum* L.) in the form of flour, starch and protein isolates in low-fat pork bologna.

In the first study, flour, starch and protein isolates from six chickpea cultivars (three Kabuli and three Desi) from two harvests (2005 and 2006) were evaluated for their physico-chemical, functional and thermal properties. Chickpea flour was made by grinding seed to pass through a 0.1mm screen, whereas protein isolates and starch were prepared by a wet milling process. Protein isolates were prepared from chickpea flour (23.2% protein on average) by alkaline extraction (pH 8.0) and isoelectric precipitation (pH 4.3). Protein isolates contained 72.8-85.3% protein; the starch fraction contained 93.0-98.0% starch. On SDS-PAGE, the chickpea flours and protein isolates contained similar polypeptide bands in the range of 30 to 55 kDa, with three major bands at approximately 50-55, 40 and 30 kDa. Least gelation concentration (LGC) for chickpea flours ranged from 6-14%; LGC for chickpea protein isolates ranged from 10-14%. Differential scanning calorimetry (DSC) of chickpea flour slurries revealed two endothermic peaks. One corresponded to starch gelatinization at approximately 64°C, which was slightly higher than for the starch fraction (~60°C). The second broad peak at approximately 96°C corresponded to the denaturation of the globulin protein fraction, which was also slightly higher than for the protein isolates (~91°C). Chickpea flour exhibited nitrogen solubility index values higher than those of chickpea protein isolates and soy and pea protein isolates. Chickpea protein isolates exhibited water holding capacities, oil absorption capacities, emulsion activity indeces and emulsion stability indeces higher than those of the

chickpea flours. CDC Xena (Kabuli) and Myles (Desi), in general, most exhibited properties appropriate for meat applications.

In the second study, the efficacy of flour, starch and protein from CDC Xena (Kabuli hereafter) and Myles (Desi hereafter) was investigated in low-fat pork bologna (LFPB). Low-fat pork bologna (<5% fat) was prepared by incorporating 2.5 or 5.0% flour, 1.5 or 3.0% protein isolate (protein basis), or 1.0 or 2.0% starch in the formulation. Controls were prepared without any binder, and formulations containing wheat or pea flour, soy or pea protein isolate, potato or pea starch, or extra meat were prepared for comparison. Inclusion of chickpea flour, protein or starch had a positive effect ($P<0.05$) on the cook yield, expressible moisture and purge of LFPB, and had little effect on colour. Increasing chickpea flour substitution from 2.5 to 5.0% altered the sensory and instrumental textural quality of LFPB significantly ($P<0.05$). Desi flour at 5.0% showed the highest TPA (texture profile analysis) hardness and chewiness, Allo-Kramer shear values and torsion shear stress. Similarly, LFPB containing chickpea protein isolate (CPI), soy protein isolate (SPI) or pea protein isolate (PPI) (3.0% protein basis) was firmer than either LFPB containing 1.5% protein from CPI, SPI or PPI or the control-I (with the same level of meat protein). Likewise, LFPB formulated with 2.0% Kabuli or Desi starch had higher TPA values than those prepared with pea or potato starch. For most flavour sensory properties, Kabuli and Desi chickpea flour and starch, irrespective of level of incorporation, performed similarly to the control. However, panellists noted more off-flavours with the addition of wheat flour or pea flour at 5.0%. Chickpea protein isolate, SPI or PPI at the 1.5% protein addition level did not alter the flavour properties of LFPB.

It was concluded that chickpea flour, starch and protein had potential for utilization as extenders in low-fat meat emulsion systems such as frankfurters and bologna.

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LIST OF ABBREVIATIONS

a*	Redness
AACC	American Association of Cereal Chemists
A-K	Allo Kramer
AM	Amit
AOCS	American Oil Chemist's Society
b*	Yellowness
CB	CDC Cabri
CDC	Crop Development Centre
CIE	International Commission on Illumination
cP	Centipoise
CPI	Chickpea Protein Isolate
CV	Coefficient of Variation
D	Desi
DSC	Differential Scanning Calorimetry
EA	Emulsifying Activity
EAI	Emulsifying Activity Index
EM	Expressible Moisture
ESI	Emulsion Stability Index
FT	CDC Frontier
g	Gram
g	Gravitational Force
G'	Storage Modulus
G"	Loss Modulus
h	Hour
HC	Hydration Capacity
HI	Hydration Index
IDF	Insoluble Dietary Fibre
Jg^{-1}	Joules per Gram

K	Kabuli
kDa	KiloDalton
kg	Kilogram
kPa	Kilopascal
L*	Lightness
LFPB	Low-Fat Pork Bologna
LGC	Least Gelation Concentration
mg	Milligram
mL	Millilitre
ML	Myles
mm	Millimetre
MW	Molecular Weight
N	Newton
N/g	Newton per Gram
nm	Nanometre
NSI	Nitrogen Solubility Index
OAC	Oil Absorption Capacity
P	Probability
PPI	Pea Protein Isolate
r	Correlation Coefficient
rpm	Revolutions per Minute
RVA	Rapid Visco Analyzer
s	Second
S	Svedberg Unit
SC	Swelling Capacity
SCT	Seed Coat
SD	Seed Density
SD	Standard Deviation
SDF	Soluble Dietary Fibre
SDS-PAGE	Sodium Dodecyl Sulphate Poly Acralamide Gel Electrophoresis
SI	Swelling Index
SPI	Soy Protein Isolate
SV	Seed Volume
SW	Seed Weight

TDF	Total Dietary Fibre
TPA	Texture Profile Analysis
USA	United States of America
VG	CDC Vanguard
w/v	Weight/Volume
w/w	Weight/Weight
WHC	Water Holding Capacity
XN	CDC Xena
δ	Loss Tangent
ΔE	Total Colour Difference
°C	Degrees Celsius
µL	Microlitre

1.0 INTRODUCTION

1.1 Rationale

Non-meat ingredients derived from a variety of plant and animal sources are used extensively as fillers, binders, emulsifiers or extenders in meat systems. These are added to reduce cost and serve as functional ingredients. Many non-meat ingredients including flours from tubers (Annor-Frempong, Annan-Prah, & Wiredu, 1996), cereals (Brown & Zayas, 1990; Salahuddin, Kondaiah, & Anjaneyulu, 1991) and legumes (Dzudie, Scher, & Hardy, 2002; Modi, Mahendrakar, Narasimha Rao, & Sachindra, 2003; Verma, Ledward, & Lawrie, 1984a), starches (Carballo, Fernandez, Barreto, Solas, & Jimenez Colmenero, 1996; Hachmeister & Herald, 1998; Shand, 2000) and proteins (Chin, Keeton, Longnecker, & Lamkey, 1999; Yang, Keeton, Beilken, & Trout, 2001) have been added to a variety of comminuted meat products. Meat batters with corn germ protein flour additives showed increased water-holding capacity (WHC) and yield and decreased cooking losses (Brown & Zayas, 1990; Wang & Zayas, 1992). Defatted corn germ protein has been reported to have high water retention, fat binding, emulsifying capacity and stability in comminuted meat products (Lin & Zayas, 1987). Wheat germ protein additives were observed to improve viscosity and adhesiveness of comminuted meat (Gnanasambandam & Zayas, 1992). Hongsprabhas & Barbut (1999) concluded that the functional and sensory characteristics of comminuted meats improved with addition of soy proteins. However, little is known about chickpea as an ingredient in meat systems.

Chickpea (*Cicer arietinum* L.) is the world's third largest pulse crop in terms of area and is grown mostly in West Asia and the Mediterranean region (Clemente, Vioque, & Sanchez-Vioque, 1998). In Canada, commercial chickpea production started in the mid-1990s and

is now widespread in Saskatchewan and Alberta. Canadian exports have followed production trends and Canada has become a major exporter of chickpea, placing among the top five in the world.

The Crop Development Centre at the University of Saskatchewan has carried out various breeding trails to scrutinize for high yielding and prairie entrenching chickpea cultivars to meet the increasing demand for chickpea (Saskatchewan Agriculture and Food, 2001). Whole chickpea contains about 17% protein, 5.3% fat, 3.0% minerals (Ravi & Bhattacharya, 2004) and 42-45% total starch (Meares, Bogracheva, Hill and Hedley, 2004). It has been reported that genotypic variation is reflected in the chemical composition of chickpea seed (Gil, Nadal, Luna, Moreno, & De Haro, 1996). Thus, a wide genetic basis may affect the functional properties of chickpea components.

Starch is a very useful raw material with many applications, from gelling in food systems such as soups, candies, meat systems, and jellies, to uses in the textile, paper and chemical industries. The growing demand for starches in the food industry has created interest in new sources of this polysaccharide, such as chickpea. Chickpea starch contains 20-30% amylose and its gelatinization temperature is between 63.5 and 69°C (Linback & Ke, 1975). Depending on its amylose content, total starch content, and crystalline structure, the specific physicochemical and functional properties of chickpea starch could vary. It is relatively easy to isolate a starch-rich fraction from chickpea by wet milling and centrifugation.

Increased interest in plant proteins in food has led to the evaluation of chickpea as a high protein crop. The protein-rich fraction obtained from different chickpea cultivars by isoelectric precipitation produced isolates with 73.0-82.0% protein content, making them excellent potential protein sources for food industry applications. This potential usefulness, however, will depend on their technologically important thermal and functional properties, which affect food sensory characteristics and play an important role in the physical behavior of food or its ingredients during preparation, processing and storage (Ahmenda, Prinyawiwatkul, &

Rao, 1999). Sánchez-Vioque, Clemente, Vioque, Bautista, & Millán (1999) investigated the chemical composition and functional properties of two types of chickpea isolates. They reported that isolates with high water and fat absorption could be suitable for the preparation of cheese or bakery and meat products, while other isolates that have high emulsion capability can be used in products such as frankfurters or creams. The functional properties and thermal properties of Indian chickpea cultivars have been reported by Kaur & Singh (2005). According to their study, protein isolates from Kabuli and Desi chickpea cultivars differed significantly in their functional and thermal properties. However, little is known of the properties of Western Canadian chickpea.

Paredes-Lopez, Ordorica-Falmir, & Olivares-Vasquez (1991) reported that micellization and isoelectric precipitation of chickpea protein isolates resulted in acceptable levels of most essential amino acids compared to the FAO/WHO/UNU reference pattern for preschool children and adults. Given the demand for new functional ingredients in the food industry, characterization of chickpea starch and protein fractions is worthwhile as it will aid in establishing its possible uses and add value to this legume seed. Hence, the present study was aimed at understanding the physico-chemical, thermal, and functional characteristics of wet-milled protein and starch fractions from chickpea in meat systems.

Many processed meats, such as frankfurters and bologna, may contain up to 30% fat. Even though fat improves the flavour and textural quality of finished products (Cross, Berry, & Wells, 1980), consumption of too much fat (>30% of total calories) in the diet may increase the risk of coronary disease and other related disorders (American Heart Association: AHA, 1978). However, reduction of fat to produce low-fat products may lead to texture and water holding problems (Claus, Hunt, & Kaster, 1989). In order to achieve favorable product characteristics when reducing fat content, functional ingredients capable of improving water binding and modifying texture are of interest to meat processors. Hence, reduced fat meat products with different functional ingredients have been studied recently (Ahn, Hsieh, Clarke, & Huff, 1999;

Andres, Garcia, Zaritzky, & Califano, 2006; Chin, Keeton, Longnecker, & Lamkey, 1998; Chin et al., 1999; Shand, 2000; Yang, Keeton, Beilken, & Trout, 2001). Utilization of chickpea in any form as an extender in low-fat meat products has not been reported. Therefore, in order to understand how chickpea fractions behave in a meat system, pork bologna was formulated with reduced fat levels using different levels of chickpea flour, starch and protein.

1.2 Objectives

The objectives of this research project were:

I. To study and compare flour and protein and starch fractions from selected chickpea varieties with respect to their physical, physico-chemical, thermal and functional properties.

II. To characterize and evaluate the impact of flour, protein and starch fractions from selected chickpea varieties on the physico-chemical, textural, cooking and sensory properties of low-fat, high moisture bologna.

2.0 LITERATURE REVIEW

2.1 Chickpea

2.1.1 Main types of chickpea

Chickpeas (*Cicer arietinum* L.) are generally grouped into two types. Kabuli chick-peas, also known as garbanzo beans, have a larger, cream-coloured seed with a thin seed coat. The Desi-type has a smaller, darker-coloured seed with a thick seed coat (Figure 2.1).

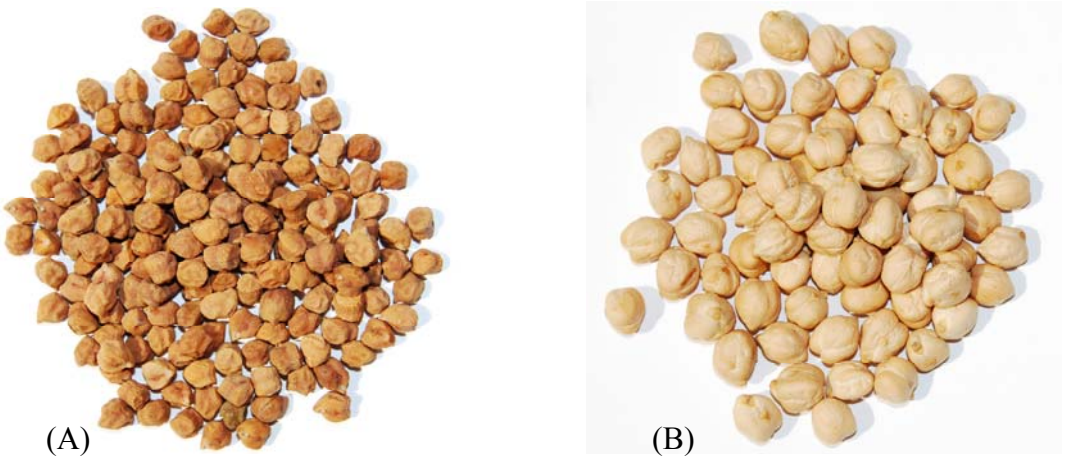

Figure 2.1 Two major chickpea lines (A) small-seeded (Desi) chickpea (B) large-seeded (Kabuli) chickpea

2.1.2 Origin and production

The cultivated chickpea, was one of the first grain legumes to be domesticated in the old world (Singh, 1997). Chickpeas are a member of the Leguminosae family, as they can fix nitrogen from the atmosphere. The growth habit is erect, with most of the pods formed in the

top part of the plant. Chickpeas most probably originated in an area of present-day South-Eastern Turkey and adjoining areas of Syria. Even though earlier botanists had postulated several different origins, Vavilov (1926) identified two primary centres of origin, south-west Asia and the Mediterranean, and one secondary centre of origin, Ethiopia. He noted that large-seeded chickpeas reached India via the Afghan capital Kabul about two centuries ago and acquired a name in Hindi as *Kabuli chana* (chana = chickpea) (Singh, 1997). The small-seeded, dark chickpea is called *Desi* (local), and these denominations are commonly used to distinguish the two main groups of cultivars. It is almost certain that small-seeded chickpeas originated first with large-seeded types developed by selection and mutation (Singh, 1997).

World chickpea production has ranged from 7-9 million tonnes in recent years, and is approximately four times larger than lentil production. However, world trade is similar to lentils because India produces and consumes approximately 4-6 million tonnes of chickpeas each year. The major chickpea exporting countries are Turkey, Canada, Australia, Syria, and Mexico (FAO, 2002).

2.2 Western-Canadian chickpea

According to Saskatchewan Pulse Growers (2006), several legumes are grown and consumed in Saskatchewan. Among these, chickpea (*Cicer arietinum*), pea (*Pisum sativum*), and lentil (*Lens esculenta*) are the major ones.

In Saskatchewan, research carried out at the Crop Development Centre, University of Saskatchewan (CDC-SK) has allowed the introduction of new varieties of chickpeas which are suitable for typical Saskatchewan weather and soil conditions. There are several Desi varieties in production namely, Myles, CDC Anna, CDC Cabri, CDC Desiray, CDC Nika, and CDC Vanguard and Kabuli varieties such as Sanford, Amit, CDC Chi Chi, CDC Chico, CDC Diva, CDC Frontier, CDC Xena, CDC Yuma, Dwellery, and Evans (Saskatchewan Pulse Growers, 2006).

Finally, although most Western-Canadian chickpeas are exported as a raw product without any processing, except cleaning and grading operations, it is important to note that increasing secondary processing activities of local chickpeas could contribute in the diversification of agriculture in Western Canada.

2.3 Proximate composition of flours from different chickpea cultivars

Chickpea cultivars grown under different environmental conditions, such as location, soil type, irrigation and fertilizers with irrigation, may have different compositions (Chavan, Kadam, & Salunkhe, 1989; Jood, Bishnoi, & Sharma, 1998; Kaur & Singh, 2005; Canadian Grain Commission, 2004). The following table (Table 2.1) summarizes the proximate and mineral composition of Canadian chickpeas (Kabuli and Desi) according to the Canadian Grain Commission (Canadian Grain Commission, 2004).

From Table 2.1 it can be seen that Kabuli has a higher content of protein (24.4% for Kabuli and 23.0% for Desi) and a wider range of protein distribution than Desi varieties. It is evident that the high level of starch content (app. 41% for Kabuli and 36% for Desi) indicates that chickpeas are an important source of easily available energy. In addition, the fibre content of Kabuli is lower (ADF 3.7% and NDF 5.0%) as compared with the fibre content of Desi (ADF 13.1% and NDF 12.8%), meaning that Kabuli chickpeas have higher degradability than the Desi-type. According to the mineral composition, chickpea is a good source of potassium and phosphorous.

Table 2.1 Proximate and mineral composition of Canadian chickpea

	Canadian chickpea (Kabuli)		Canadian chickpea (Desi)	
	Mean	Range	Mean	Range
Composition (g/100 g dry matter)				
Protein (N×6.25)	24.4	17.9-30.8	23.0	20.3-27.5
Starch	41.1	38.2-43.9	36.4	33.1-40.4
Amylose (% of total starch)	26.2	24.4-29.2	23.8	20.5-25.9
ADF[a]	3.7	3.0-5.7	13.1	12.7-13.5
NDF[b]	5.0	4.2-7.7	12.8	10.1-13.6
Fat	5.9	5.5-6.9	5.4	4.4-5.9
Ash	3.2	2.9-3.8	3.2	2.7-3.5
Minerals (mg/100 g dry matter)				
Calcium (Ca)	106.6	80.5-144.3	161.7	115.0-226.5
Copper (Cu)	1.0	0.7-1.4	1.0	0.5-1.4
Iron (Fe)	5.5	4.3-7.6	5.9	4.6-7.0
Potassium (K)	1127.2	816.1-1580.1	1215.7	1027.6-1479.1
Magnesium (Mg)	177.8	152.9-212.8	169.1	143.7-188.6
Manganese (Mn)	3.9	2.3-4.8	3.4	2.8-4.1
Phosphorus (P)	505.1	294.1-828.8	377.3	276.2-518.6
Zinc (Zn)	4.4	3.6-5.6	3.6	2.8-5.1

ADF[a] = acid detergent fibre, and NDF[b] = neutral detergent fibre. (Modified from Canadian Grain Commission, 2004)

2.4 Preparation of chickpea flour, starch-rich and protein-rich fractions at a lab scale

2.4.1 Flour preparation

Chickpea flour can be obtained by milling. Tabil & Mani (2002) have reported a pin mill method for chickpea flour production. Seeds are passed through a pin mill. Pin milling breaks up the seed by impact and converts seeds to a fine flour. A pin mill consists of a number of short pegs or pins in two discs in a concentric fashion. The discs are powered by high-speed motors, rotating at a speed of 34,000 rpm in opposite directions. Chickpea flour produced from a pin-mill has an average particle size of 0.18 mm (Emami & Tabil, 2002).

The chickpea also can be ground using a turbo mill. The turbo mill R9000 (RETSCH SR300, Haan, Germany) fitted with 250-micron screen, consisting of an electromagnetic vibratory feeder, high-speed beater and a screen has been used by Emami & Tabil (2002). A high size reduction was achieved through a combination of impact, rebound and shear action by the high-speed of the rotor beater (8100 rpm). The turbo-milled chickpea flour also had similar particle size as pin-milled chickpea flour (Emami & Tabil, 2002).

Costa, Oueiroz-Monici, Reis, & Oliveira (2006) used a combination of hydration and thermal treatment to obtain flour from grains. In this method, chickpeas are soaked for a period of 16 h (1:2 w/v) and then cooked with the addition of one volume of water in a domestic pressure cooker (14.7 *psi*) for 40 min. The cooked material was frozen, freeze-dried (Virtis, 10-146 MR-BA model) and ground into flour (#60 mesh).

2.4.2 Preparation of starch- or protein-rich fractions

Separation of starch- or protein-rich fractions from chickpea flour can be obtained through either dry milling or wet processing.

2.4.2.1 Dry processing - air classification

Air classification is often used to produce protein or starch concentrates from cereals and pulses (Tyler, Youngs, & Sosulski, 1980; Vose, Basterrechea, Gorin, Finlayson, & Youngs, 1976). Flour particles produced by pin milling are different in their shape, size and density. Air classification differentiates the protein (fine fraction) and starch (course fraction) particles. The particles can be air classified in a centrifugal or gravitational (crossflow or counterflow) air stream, and can then be fractionated into light and heavy particles (Shapiro & Galperin, 2005).

The pin milled flour is air classified in a spiral air stream and fractionated into light and heavy particles. The starch-rich fractions contain 58-76% starch and 8-20% protein (depending on the legume source). The protein-rich fractions contain 49-75% protein and 0-5% starch. If starch fractions are remilled and air classified again, the recovery of the starch fraction and the protein fraction will improve. The starch fractions in the second stage contain 71-86% starch and 4-10% protein whereas; the protein fractions contain 38-68% protein and 0.4-1.7% starch (Tyler, Youngs, & Sosulski, 1980).

Meares et al. (2004) used a zigzag air classifier A 100MZR to separate chickpea flour into starch- and protein-rich fractions. The speed of the air classifier motor was 7000 rpm, the airflow was 48 m^3/h and the feed rate was 0.7 kg/h. The starch-rich fraction had a total starch content of ~ 46% and a protein content of ~ 20%. The protein-rich fraction had a starch content of ~ 40% and a protein content of ~ 24%.

2.4.2.2 Wet processing

Wet processing is employed to prepare highly purified protein and starch fractions. However, a higher amount of energy is spent for drying and refining of the effluent in wet processing, making it difficult and costly. Protein concentrates and protein isolates (high protein concentration) from pulse grains can be prepared by wet processing. There are four well-known procedures in practice under this method, namely, isoelectric precipitation method, salting out method, solvent extraction, and ultrafiltration method (Nash, Eldridge, & Wolf, 1967; Wu, 1993; Chau, Cheung, & Wong, 1997; Sánchez-Vioque et al., 1999; Sze-Tao, & Sathe, 2000).

2.5 Colour and functional properties of chickpea

2.5.1 Colour characteristics

Determination of colour for each chickpea variety is very important since it may play a vital role in the end product, especially in meat products. A Hunterlab colourimeter could be used for measuring colour of chickpea varieties on the basis of CIE L^*, a^*, b* and ΔE values. Kabuli-type chickpea flour from Indian cultivars showed the highest L^* and ΔE value, indicating its lighter colour than flours from Indian Desi-type chickpea cultivars (Kaur & Singh, 2005; Kaur, Singh, & Sodhi, 2005).

2.5.2 Water holding capacity (WHC) and oil absorption capacity (OAC)

Water absorption characteristics represent the ability of a product to associate with water under conditions where water is limited. Kabuli chickpea flours (1.37-1.47 g/g) showed a significantly higher WHC than Desi-type flour (1.33 g/g) (Kaur & Singh, 2005). The higher water absorption of Kabuli chickpea flours compared to Desi chickpea flours could be attributed to the presence of hydrophilic constituents such as polysaccharides and proteins in them (Kaur & Singh, 2005). The WHC of chickpea starches has also been investigated (Patanè,

Iacoponi, & Raccuia, 2004; Singh, Kaur, Sandhu, & Guraya, 2004; Singh, Sandhu, & Kaur, 2004). The WHC of different chickpea cultivars ranges between 77.8-89.4%.

The oil absorption capacity (OAC) of flours is also important, as oil improves the mouth feel and retains flavour of final product (Kinsella, 1981). Kabuli chickpea flour showed significantly higher OAC (1.24 g/g) than the OAC of Desi chickpea flours (ranged from 1.05 to 1.17 g/g) (Kaur & Singh, 2005). According to Kinsella (1981), proteins that are more hydrophobic show superior ability to bind of lipids, implying that non-polar amino acid side chains bind the aliphatic chains of fats. Based on this suggestion, it could be inferred that Kabuli chickpea flour, which showed a higher OAC, has more non-polar side chains in its protein molecules than Desi chickpea flours.

2.5.3 Emulsifying activity

Emulsifying activity (EA) is defined as the ability of the flour to emulsify oil. Flours from various chickpea cultivars differ significantly in their ability to emulsify fat. According to Kaur & Singh (2005), flours from Kabuli chickpea cultivars showed significantly lower emulsifying ability (58.2%) than did Desi chickpea flours (59.6-68.8%). The difference in total protein composition (soluble and insoluble), as well as components other than proteins (possibly carbohydrates), may contribute substantially to the emulsifying properties of protein-containing products like legume flours. Furthermore, the factors affecting emulsification are related to the physico-chemical characteristics of the proteins – surface hydrophobicity and charge, steric effects, elasticity-rigidity, viscosity in solution, and the ability of the macromolecules to rearrange after absorbing at the interface and to form a continuous film of high mechanical strength (Sikorski, 2001).

2.6 Thermal properties

2.6.1 Differential scanning calorimetry

Thermal stability of chickpea flour, starch and protein can be determined by using differential scanning calorimetry (DSC). DSC monitors changes of polymers (carbohydrates, proteins, or lipids) associated with phase transitions and chemical reactions as a function of temperature. DSC can be used to detect both first-order (melting) and second-order (glass transition) transitions. In food research, first-order transitions are mainly related to processes such as protein denaturation, starch gelatinization and fat crystal melting (Ma, Harwalkar, & Maurice, 1990). Using DSC, the following are commonly determined:

Tg = glass transition temperature, the temperature (°C) at which an amorphous polymer or an amorphous part of a crystalline polymer goes from a hard brittle state to a soft rubbery state.

Tm = melting point, the temperature (°C) at which a crystalline polymer melts.

ΔHm = the amount of energy (joules/gram) which a sample absorbs while melting.

Tc = crystallization point, the temperature at which a polymer crystallizes upon heating or cooling.

ΔHc = the amount of energy (joules/gram) a sample releases while crystallizing.

In the case of a food sample, the DSC curves give information on thermal properties such as onset (T_o), peak (T_p), conclusion (T_c), peak height index (PHI) and gelatinization/denaturation range ($T_c - T_o$).

Thermal characteristics (gelatinization temperatures) of flours from different chickpea cultivars have been studied by using a differential scanning calorimeter equipped with a thermal analysis data station (Kaur & Singh, 2005; Meares et al., 2004; Singh et al., 2004).

Literature findings for the thermal properties of chickpea flour and starch are shown in Table 2.2. Generally, peak temperature of chickpea flour samples was slightly higher than for the equivalent pure starch sample. Desi starch had the highest values of T_o, T_p and T_c whereas Kabuli starch showed lowest values for the same (Linback & Ke, 1975). The differences in gelatinization temperature may be attributed to the differences in amylose content, size, form and distribution of starch granules, and to the internal arrangement of starch fractions within the granules (Singh, Sandhu, & Kaur, 2004).

Table 2.2 Thermal characteristics of chickpea flour and starch

Chickpea fraction	DSC parameter*				Reference
	T_o (°C)	T_p(°C)	T_c(°C)	ΔH_{gel}(J/g)	
Flour	64.0	72.0	-	0.74	Meares et al. (2004)
	65.4-67.9	70.6-73.3	77.0-79.4	3.5-4.9	Kaur & Singh (2005)
Starch	62.0	69.0	-	0.78	Meares et al. (2004)
	61.5-64.8	66.4-69.0	71.3-73.8	7.2-8.7	Singh et al. (2004)
Kabuli-starch	60.0	65.0	75.0	-	Linback & Ke (1975)
Desi-starch	63.5	65.0	69.0	-	Linback & Ke (1975)
	59.4-59.7	64.7-67.7	71.7-78.2	9.7-12.4	Hoover & Ratnayake (2002)

* T_o; onset temperature, T_p; peak temperature, T_c; conclusion temperature, ΔH_{gel}; enthalpy of starch gelatinization

One of the most useful characteristics of starch is the enthalpy of starch gelatinization (ΔH_{gel}). ΔH_{gel} can be calculated as the area under the gelatinization peak. Kaur & Singh (2005) reported ΔH_{gel} value of flours from different chickpea cultivars ranging from 3.5 to 4.9 J/g, the highest for Desi-type and lowest for Kabuli-type.

Tester (1997) postulated that the gelatinization and swelling properties are controlled in part by the molecular structure of amylopectin (perfection and ordering of amylopectin crystallites, length of the external 'A' chains of amylopectin, extent of branching, molecular weight

and polydispersity), starch composition (amylose/ amylopectin ratio, lipid complexed amylose chains) and granule architecture (crystalline to amorphous ratio). Cooke & Gidley (1992) showed that ΔH_{gel} reflects loss of double helical order rather than the loss of crystallinity. However, Tester, Morrison, & Schulman (1993) postulated that ΔH_{gel} reflects the overall crystallinity (quality and amount of crystallites) of amylopectin.

According to Singh et al. (2004), there are positive relationships between ΔH_{gel} and peak height index (PHI) of starches from different chickpea cultivars ($r^2=0.6189$). PHI is the ratio of ΔH_{gel} for gelatinization to the gelatinization temperature range and is a measure of uniformity in gelatinization. Kaur & Singh (2005) reported PHI values for various chickpea flours ranging from 0.67 to 0.93. Kabuli-type cultivar showed the significantly lower T_o (65.4°C), T_p (70.6°C), T_c (77.0°C), ΔH_{gel} (3.5 J/g), and PHI (0.67) as compared to Desi chickpea flours. So, less energy is needed (fusion enthalpy) to break the intermolecular bonds in starch granules of Kabuli flours to achieve gelatinization.

Nevertheless, there is no published gelatinization information for Saskatchewan grown chickpea cultivars. Therefore, it is important to study thermal properties of chickpea flour, starch and protein to understand thermal behavior of chickpea components.

2.6.2 Pasting properties of starches

2.6.2.1 Viscosity profile

A Rapid Visco Analyser (RVA) is widely used to obtain a viscosity profile or visco-amylograph of starches. RVA is becoming popular, because it has the advantages of using a small sample size, short testing time, variable bowl speed and electronic recording of the results. According to Thomas & Atwell (1997), there are several factors such as type of starch, solid levels, pH of the slurry, and heating regime that influence various gelatinization or pasting profiles.

A typical Rapid Visco Analyzer profile from normal maize starch is shown in Figure 2.2. A pictured illustration also indicates granular changes along the curve. These starches showed a gradual increase in viscosity with the increase in temperature. Granule swelling is accompanied by leaching of granular constituents, predominantly amylose, into the external matrix resulting in a dispersion of swollen granules in a continuous matrix (Tester, Karkalas, & Qi, 2004).

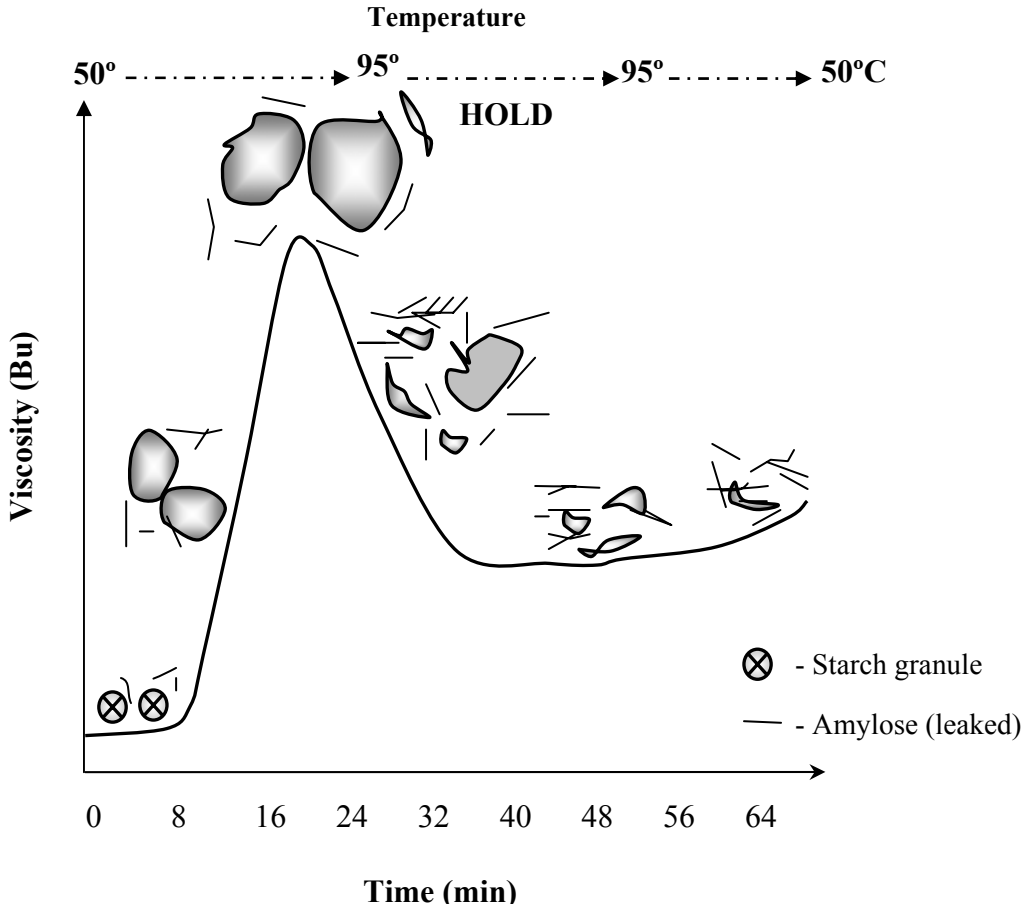

Figure 2.2 Schematic representation of granular changes in relationship to viscosity. The viscosity profile was measured by a viscoamylograph (Modified from Thomas & Atwell, 1997).

In this RVA profile, native starch granules are generally insoluble in water below 50°C. Thus, the viscosity is low. Then as temperature increases, granules begin to swell to many times their original size because of water absorption. Lii, Shao, & Tseng (1995) reported that

rice starch granules, before gelatinization, swelled two to four times. The peak viscosity is reached when most of starch granules are swollen. As the temperature increases further and is held at a high temperature (e.g., 95°C) for a period of time, granules rupture and subsequent starch polymer alignment occurs, which decrease the apparent viscosity of the paste. This process is known as breakdown and it gives an idea of stability of the starch paste. It is important to stress that only intact swollen granules give paste viscosity, and not fragmented granules or solubilized polymers (Thomas & Atwell, 1997).

Upon cooling, the amylose molecules aggregate, leading to gel formation (Schiraldi, Piazza, & Riva, 1996). Thus, the viscosity increase on cooling is a measure of the retrogradation tendency of the starch. This phase of the pasting curve is commonly referred to as the setback region.

2.6.2.2 RVA applications in chickpea studies

The pasting properties of the chickpea flour, starch and protein rich fractions measured using a RVA have been reported. Singh et al. (2004) reported pasting temperatures of starches from chickpea cultivars ranging between 75.1-77.1°C. Linback & Ke (1975) reported pasting temperature of 68.5°C for chickpea starch and 67°C for horse bean starch. Pasting temperature of 66°C for faba bean and 71°C for mung bean starches have been reported earlier by Naivikul & D'Appolonia (1979). Singh et al. (2004) reported pasting temperature between 75.8-80.3°C for black gram cultivars. High pasting temperature of chickpea and black gram starches indicated their high resistance towards swelling.

According to Kaur & Singh (2005), flour from Indian grown Kabuli chickpea cultivars had a lower pasting temperature (73.9°C), highest peak viscosity (180.3 RVU), trough (162.5 RVU), final viscosity (225.3 RVU) and setback (62.8 RVU), while Desi chickpea flours had the highest pasting temperature (75.2°C), lowest peak (112.3 RVU) trough (106.4 RVU), breakdown (5.9 RVU), final viscosity (126.3 RVU), and setback (19.8 RVU). Hence, the re-

sults revealed that chickpea flour or starch is suitable for food uses where a thermo-stable paste without breakdown and with restricted swelling is required.

Pasting temperature provides an indication of the minimum temperature required to cook the flour. When chickpea flour is heated in water, starch gelatinization occurs, followed by protein denaturation (Meares et al., 2004). The pasting properties of chickpea flour are considerably lower than that of the chickpea starch. Sayar, Koksel, & Turhan (2005) indicated that the addition of protein-rich fraction of chickpea to the starch substantially decreases the pasting properties of the starch. A previous study by Fitzgerald, Martin, Ward, Park, & Shead (2003) carried out with rice flour, indicated that the denatured proteins stabilize the continuous matrix or strengthen the links between the dispersed and continuous phases. They also stated that proteins offer some protection against breakdown, which is further supported by the greater lift-off observed when proteins are present.

The pasting behavior of chickpea flour/starch can be changed by the addition of different additives such as salt, oil, and sodium carboxymethyl cellulose (Ravi & Bhattacharya, 2004) as well as removing substances such as fat and protein from chickpea flour/starch.

2.6.3 Dynamic oscillatory rheology measurement

2.6.3.1 Principle and application

Rheological measurements determine the relation between the deformation (strain) of a material and the stress applied to achieve such a deformation as a function of time. They can be performed at small and large deformations. At the small deformation, the measurement is performed in such a way that the microstructure of the gel is not damaged and that the obtained modulus is independent of the applied strain (*i.e.* in the linear region). In large deformation measurements, the gels are mostly deformed until macroscopic fracture takes place. Small deformation measurements are often performed using dynamic mechanical rheology, in which the applied strain (or stress) varies sinusoidally. Parameters obtained from dynamic measurements

are the storage or elastic modulus G', which is a measure of the amount of energy that is stored during a periodic application of stress or strain, the loss or viscous modulus G'', which is a measure of the energy loss, and their ratio $tan\ \delta = G''/G'$, which is called the loss tangent. With dynamic measurements, the onset of gel formation can be determined and the stiffening of the gel can be followed. Parameters obtained in large deformation or fracture measurements are stiffness (Young's modulus), fracture stress, fracture strain, and fracture energy (Walstra & Van Vliet, 1992). These mechanical properties of gels are the most important ones for practical reasons, *i.e.* during handling, slicing and eating of the gels.

Rheological properties of plant protein gels have been extensively studied, including viscoelasticity or gelation of 7S globulin from soybeans (Nagano, Hirotsuka, Mori, Kohyama, & Nishinar, 1992), monitoring of gluten/soy protein gelation with heat treatment (Apicharts-rangkoon, 2002), thermal gelation of the 12S canola globulin (Leger & Arntfield, 1993), and thermal gelation of globulin from red bean (*Phaseolus angularis*) (Meng & Ma, 2002), pea legumin (O'Kane, Happe, Vereijken, Gruppen, & Van Boekel, 2004), vicilin (7S protein) from faba bean (Arntfield, Murray, Ismond, & Bernatsky, 1989; Arntfield, Murray, & Ismond, 1990a; Arntfield, Murray, & Ismond, 1991; Arntfield, & Murray, 1992) and chickpea protein isolates (Zhang, Jiang, & Wang, 2007).

2.7 Meat applications of plant-base ingredients

Plant based ingredients, mainly from legumes and cereals, have been used as binders and extenders in comminuted meat products. There are two main reasons for the usage of plant ingredients in meat products; first, to reduce the price of the products and secondly, to enhance the functionality. Plant proteins are used in meat products to perform three basic functions: the first function is fat emulsification, the second is water retention, and the third is formation of structure of meat products. Different non-meat ingredients, which have been used in different meat systems, are summarized in Table 2.3.

Table 2.3 Plant ingredients incorporated in different meat products

Meat system	Plant Ingredient	Reference
Low-fat bologna	Soy protein isolates and konjac blends, normal or waxy starch hull-less barley	Chin et al. (1998); Shand (2000)
Fresh sausage	chickpea flour	Verma, Ledward, & Lawrie (1984a,b, 1987)
Cured/emulsified sausage	Common bean flour	Dzudie, Scher, & Hardy (2002)
Low fat meat balls	chickpea, bean, lentil and rusk	Serdaroğlu, Yıldız-Turp, & Abrodímov (2005)
Meat burgers	soybean, faba bean, chickpea, Bengal gram, black gram and rice flour	Modi,, Mahendrakar, Narasimha Rao, & Sachindra (2003); Moharram, Hamza, Aman, & El-Akary (1987)
Chicken nuggets	cowpea and peanut flour	Prinyawiwatkul, McWatters, Beuchat, & Phillips (1997)
Beef patties	corn germ protein flour	Brown & Zayas (1990)
Comminuted meat products	wheat germ protein, soya proteins	Gnanasambandan & Zayas (1992); Lecomte, Zayas, & Kastner, (1993); Kassama, Ngadi, & Raghavan, (2003); Rentfrow, Brewer, Weingarartner, & McKeith, (2004)
Ground beef mixture	wild rice	Minerich, Addis, Epley, & Bingham (1991)
Buffalo meat loaves	*Detarium microcarpum* seed flour	Onweluzo, Puttarajappa, Sakhare, & Narasimha Rao (2003)
Meat batters	wheat gluten, soy protein isolates,	Chen & Trout (1991)
Reduced-fat turkey batters	acid thinned dent corn, cross-linked waxy maize, cross-linked dent corn, modified tapioca and modified potato starches	Hachmeister, & Herald (1998)

Moharram et al. (1987) reported that addition of plant ingredients such as soybean, faba bean, chickpea, and white rice flour led to an increase in the levels of moisture by 12-34% and

carbohydrates by 7-29% and to a decrease in total protein of 9-24%, fat 3-5% and ash 1-5% except in the case of soybean addition, which caused a noticeable rise in ash content. Also, slight differences were noticed between the amino acid contents of meat containing different types of plant meat substitutes. These results agree with those reported by Brown & Zayas (1990) with addition of corn germ protein flour, by Dzudie, Scher, & Hardy (2002) with addition of common bean flour into the beef patties, by Minerich et al. (1991) with increased level of added wild rice flour into ground beef mixtures and by Prinyawiwatkul et al. (1997) with addition of cowpea and peanut flours to chicken nuggets. An additional factor is a dilution effect in the protein when the flour additive was hydrated with water (three times the ingredient's weight). Therefore, the reduction in protein content resulted in a higher retention of water and fat in batters extended with flour.

However, several researchers have found that protein and moisture contents of meat batters increased with addition of soy proteins (Gnanasambandam & Zayas, 1992; Lecomte et al., 1993) and soy, black gram and green gram flours (Modi et al., 2003).

2.7.1 Low-fat meat products

Processed meats, such as frankfurters and bologna, may contain up to 30% fat. Fat in these products is important due to its contribution to flavor and texture (Cross et al., 1980). Fat also serves as a source of essential fatty acids, which are precursors for prostaglandins and essential components of cell membrane. However, consumption of too much fat (>30%) in the diet has been reported to increase risk of coronary heart disease and other related disorders (AHA, 1978). Emphasis is placed on reducing total dietary fat and meat processors have responded with low-fat meat products (LFMP) (Ahn et al., 1999; Andres et al., 2006; Chin et al., 1998; Chin et al., 1999; Shand et al., 1990; Yang et al., 2001). Any combination of added water and fat may be used in cooked sausages that do not exceed a combined total of 40% and the fat content may not exceed 30% (USDA, 1990).

Fat plays an important role in the formation of stable meat emulsions (Hughes, Confrades, & Troy, 1995), while some low-fat versions of sausage, for example, may be perceived as having a less acceptable texture. Hence, there are a number of issues to be resolved in order to produce low-fat products that are acceptable to a given market. For example, reduction of fat content below 20% (generally by substituting water for fat) in meat products can lead to problems such as unacceptable texture, flavour, and appearance (Yang et al., 2001). Fat replacements or substitutes are ingredients that contribute a minimum of calories to formulated meats and do not dramatically alter flavor, juiciness, mouthfeel, viscosity or processing properties. Fat has been replaced in processed low-fat meat products with added water, nonmeat proteins, polysaccharide gums and other compounds such as polydextrose (Chin et al., 1998; Chin et al., 1999).

Legumes, particularly soybeans, have been the most extensively used for as fat replaces and water binding ingredients (Williams & Zabik, 1975). Gum and starches have also been used as partial fat substitutes (Onweluzo et al., 2003). In 1971, the USDA's food and nutrition workers recommended the use of up to 30% hydrated textured vegetable products to replace up to 30% of the meat (Williams & Zabik, 1975). The replacement of fat with these substitutes is an attractive approach in low-fat meat products since these ingredients can maintain the functional properties of the product while imparting fat-like properties.

2.7.2 Proximate composition of comminuted meat products

The nutritive value of all foods, including meat and meat products, is being seriously considered in view of consumer interest and demand. Nutritional labeling is now required for most manufactured meat products. The major meat packers and processors are proving information on the nutritional value of most products. According to the 1990 Canadian Meat Inspection Regulations, Canadian standardized meat products such as sausage (ready to eat) including, salami, weiners, frankfurters and bologna are required to contain a minimum protein

content derived from meat (9.5%) and total protein (11%). The minimum meat protein content is described as per a cooked meat weight basis unless specified otherwise. Unlike U.S. meat products, both red meat and poultry products are controlled in a similar manner. Hence it is necessary to analyze proximate composition of new products. Grossly speaking, meat is composed of water, fat, protein, mineral (ash), and a small proportion of carbohydrates, but the composition may change with addition of other ingredients such as salt, extenders, binders and fillers.

2.7.3 Colour of comminuted meat products

Appearance is an important consumer requirement, colour being the most important attribute. Natural meat colour is due to the combined effects of red pigments in muscle (myoglobin and hemoglobin) blended into the other meat components. In other words, the percentage of myoglobin and hemoglobin in combination with muscle, fat, iron and connective tissue determines meat colour (Aberle, Forrest, Gerrard, & Mills, 2001).

Unusual colour development may occur in meat in several ways, some of which are unrelated to normal chemical reactions of the pigments. In the case of meat blended with other ingredients, these ingredients may influence the colour of the final product.

For measuring colour instrumentally, the meat batters are often evaluated for L^* (lightness), a^* (redness), and b^* (yellowness) values using a CIE colour measurement. The data are then used to calculate the indices of hue and saturation as follows:

$$\text{Hue angle: } H = \tan^{-1} b^*/a^*, \qquad [2.1]$$

$$\text{Saturation index: } S = (a^{*2} + b^{*2})^{1/2} \text{ (Little, 1975)} \qquad [2.2]$$

Verma et al. (1984a, 1984b) reported that during the preparation of sausages containing chickpea flour, brown discolouration of the batter was observed at the mixing stage. Chickpea flour itself does possess a yellowish tinge, but subjective and objective assessment indicated

that the browning was not related to the inherent colour of the flour itself but appeared to be related to the conversion of the heamatin pigments present in the meat (mainly myoglobin) to the brown oxidized form. Verma et al. (1984a) also postulated that in the presence of unsaturated fat and lipoxidase (chickpea flour is rich in unsaturated fats and also contains the fat oxidizing enzyme, lipoxidase), marked acceleration of metmyoglobin formation does occur, presumably via reaction with the products of the oxidizing lipids. They also demonstrated that the increased rates of lipid and myoglobin oxidation occurring in the sausages containing chickpea flour can be prevented by appropriate heat treatment of the flour to inactivate the enzyme, lipoxidase. Similar findings were observed by Prinyawiwatkul et al. (1997) when chicken nuggets were extended with cowpea and peanut flour. They reported that as the amount of cowpea and peanut flour increased, a more intense orange-brown colour was reported (low L*, b* and hue angle). There were no differences in a* values of other treatments. Colour values of the sausages with and without added common bean flour (CBF) were studied by Dzudie et al. (2002). They reported that samples containing CBF were significantly lighter (L*-value) ($P<0.05$) than the control sample except for the sample with CBF at level of 2.5% CBF. Lecomte et al. (1993) reported that incorporation of soy proteins in pre-emulsified fat or as powders in frankfurter formulation had no detrimental effect on colour.

2.7.4 Water holding properties

2.7.4.1 Water-holding capacity (WHC) of comminuted meat products

The ability of meat to retain naturally occurring or added water during application of external forces such as cutting, heating, grinding, or pressing is highly variable. Some loss of moisture occurs even during the mildest application of these treatments because a portion of the water present is in the free form. Many of the physical properties of meat (including colour, texture, and firmness of raw meat, and juiciness and tenderness of cooked meat) are partially dependent on water-holding capacity.

Water-holding capacity is especially critical in meat ingredients of meat products that are subjected to combinations of heating, grinding, and other processes. Weight losses during fabrication processes are largely the result of water evaporation. Nevertheless, the proper protein/water ratio is important for palatability and adequate yield of the finished product. Serdaroğlu et al. (2005) reported that meatballs extended with legume (including chickpea) flours had higher WHC than meatballs extended with rusk. Lecomte et al. 1(993), Gnanasambandam & Zayas (1992), Hongsprabhas & Barbut (1999) reported that different proteins, such as soy, corn germ, whey, wheat germ, when incorporated as extender increased the WHC. Dzudie et al. (2002) reported that the addition of common bean flour increased WHC of beef sausages. Hongsprabhas & Barbut (1999) observed that using preheated whey protein isolate was very beneficial in increasing WHC, reducing cook loss, and increasing gel strength of raw and cooked meat products, particularly at low salt levels. Hung & Smith (1993) observed that whey protein play an important role in the gel formation with chicken breast salt soluble protein. They observed that characteristics of globular structures of whey protein are still present at high temperature while the typical salt soluble meat protein network was not observed in the microstructure. These microstructures suggest the importance of whey protein gelation in combination gels heated above the denaturation temperature of β-lactoglobulin (the major whey protein).

Apart from functional ingredients mentioned above, there are several other factors, which influence the WHC of meat products. The addition of brine (NaCl) also helps to form a more stable meat matrix, which leads to a smaller release of water and fat thus improving binding properties of restructured meats (Carballo, Mota, Barreto, & Colmenero, 1995; Pietrasik & Shand, 2003). Moiseev & Cornforth (1997) reported that the binding strength and cohesiveness of restructured beef rolls depend on the added water content and pH of meat.

2.7.4.2 Expressible moisture (EM)

A range of approaches for determining EM have been made. Expressible moisture refers to the amount of liquid squeezed from a protein system by the application of force and measures the amount of loose water released under the measurement conditions. No absolute figures exist since the amount of loose water depends upon the type and amount of force applied (Jauregui, Regenstein, & Baker, 1981). A simple method of measuring expressible moisture was described which combines a gravimetric adaptation of the filter paper press method (Karmas & Turk, 1976) with the application of a centrifugal force (Miller, Saffle, & Zirkle, 1968).

2.7.4.3 Purge

Even though this method is hard to categorize as a water holding method, it gives valuable information about the water release rate of the finished product during prolonged times of storage ranging from 14 days (Carballo et al., 1995) to 14/28 days (Shand, 2000), to 30 days (Gregg, Cluas, Hackney, & Marriot, 1993), to 9 weeks (Beggs, Bowers, & Brown, 1997). Furthermore, purge from vacuum packaged product could be considered a form of expressible moisture due to the combined effects of gravity and the decreased partial pressure in the package (Shand, 1999).

2.7.5 Cooking yield

Cooking improves meat palatability by intensifying the flavour and altering texture. It destroys considerable numbers of microorganisms improves the storage life of meat products, inactivates endogenous proteolytic enzymes and prevents development of off-flavours. Moreover cooking decreases the water content of raw meat, especially on the surface, which in turn lowers the water activity and improves the peelability of frankfurters and extends their shelf life. Cooking also helps to stabilize the red colour in cured meats, and it modifies the texture or

tenderness of meat and meat products (Pearson & Gillett, 1999). On the other hand, the biggest problem with cooking is it reduces yield of final products. Cooking yield can be determined as the percentage difference between the fresh batter and the cooked weights.

Verma et al. (1984b) reported marked decreases in total cooking yield in sausages (made out of mutton, pork, and beef) as the substitution (chickpea flour) level increased ($P<0.001$). But contrary findings were reported by Moharram et al. (1987). They reported that the addition of rice, faba bean, chickpea, and soybean led to a reduction of both weight loss and shrinkage of meat product. Similarly, cooking yield increased with an increased amount of common bean flour in the beef sausages formulations (Dzudie et al., 2002) and the addition of corn germ protein flour (CGPF) increased WHC and cooking yield of CGPF-extended beef patties over that found in the control beef patties (Brown & Zayas, 1990).

There are several factors that contribute to cooking losses. Sheard, Nute, & Chappell (1998) and Serdaroğlu et al. (2005) reported that loss of fat content in meat products during cooking is one of the main reasons for lower cooking yield. Mortensen, Andersen, Engelsen, & Bertram (2006) stated that pH, freezing temperature, cooking rate, and water distribution within the meat products are the main reasons for the cooking losses. Whereas, Brewer & Novakofski (1999) reported that cooking rate had no effect on cooking loss of lean ground beef. Anderson & Berry (2001) observed that 10% fat containing beef patties extended with pea fibre had higher fat retention and higher cooking yield compared to the all-beef controls. However Troutt et al. (1992a, 1992b) observed higher cooking yields but no change in fat retention in 5% fat containing patties with the use of sugar beet fibre, oat, fibre, and polydextrose.

2.7.6 Gelation

2.7.6.1 Gelation of meat protein

Solubilization of myosin with salt and water and further unfolding of the protein by exposure to fat during emulsification helps to produce a protein network or gel matrix (Pearson &

Gillett, 1999). Factors affecting gel characteristics include pH, ionic strength, protein extractability, amount of connective tissue, and heating conditions. Modifying these properties can change the microstructural and viscoelastic properties of the meat products (Aguilera & Stanley, 1993). In meat gels (heat induced), there are different types of interactions including protein-water interaction, protein-fat interaction and protein-protein interaction (Acton & Dick, 1989) which consist of multiple hydrogen bonds (Eldridge & Ferry, 1954), electrostatic and hydrophobic interactions (Wolf & Tamura, 1969), disulfide bonds (Huggins, Tapley & Jensen, 1951) or peptide bonds (Bello, 1965).

2.7.6.2 Gelation of plant-meat protein mixtures

Non-meat ingredients such as plant proteins may be used in meat products to alter the type of gel formed. In plant protein, globulin is the major protein responsible for the gelation. To optimize the use of non-meat proteins in meat products for desirable functional properties, it is necessary to understand the interactions that may occur between meat proteins and the added protein during processing. Such ingredients are used to improve yield, modify textural properties, and reduce costs of various meat formulations (Hung & Smith, 1993). Meat products are usually cooked to a final internal temperature of 73°C. Muscle proteins heated to this temperature are essentially fully denatured allowing the exposed reactive groups to impart desirable functionalities in processed meats (Liu, Xiong and Butterfield, 2000). Nonmuscle proteins may be dispersed in the muscle meat gel matrix to bind water, or they may gel and thus interact with the muscle proteins (Foegeding and Lanier, 1987) to form multicompenent gels (Tolstogusov & Braudo, 1983).

However, further, there have not been any reports on gelation of other plant proteins including chickpea in meat systems.

2.7.7 Texture / tenderness

2.7.7.1 Application

Cooking meat always leads to textural changes of meat products. Starting at 65°C (149°F), muscle (myofibrillar) protein begins to harden and become less tender (tough) (Romans, Costello, Carlson, Greaser, & Jones, 1985). Collagen begins to become solubilized (tender) in this temperature range. Thus, cuts that have low amounts of connective tissue should not be heated (cooked) beyond this temperature because increased heating will toughen them. The other factors responsible for textural properties in comminuted meat products are mainly the degree of extraction of myofibrillar protein, stromal protein content, degree of comminution and type and level of non-meat protein (Romans et al., 1985).

Dzudie et al. (2002) reported that addition of common bean flour (CBF) at 5.0, 7.5 and 10.0% of the weight of meat to beef sausages had significant effects on textural properties. It lowered the shear force and hardness of sausages compared to that of control ones. They also postulated that the substitution of CBF for muscle dilutes the quantity of connective tissue in CBF-extended beef sausages and accounted for lower shear force values. Cohesiveness of sausages was higher than that of control sausage when CBF was added at the level of 5.0 to 10% of the formulation of beef sausages. Similar results were obtained in the work of Chen & Trout (1991) who found that restructured beef steak with soy protein isolate had a higher score for cohesiveness than control beef steak.

The addition of starch into comminuted meat products has been widely investigated. Starch influenced the microstructure of meat emulsions. Usually, the starch in the emulsion is generally gelled, located inside cavities, and covering the interior walls, which are in contact with the matrix surrounding the cavities (Couso et al., 1994). The origin of the starch can affect the properties imparted to meat products. Waxy cornstarch, which principally consists of amylopectin (branched chained), has a natural tendency to give a more fat-like feel than dent

corn starch, which has 74% amylose (straight chained) and only 26% amylopectin chains (Pearson & Gillett, 1999). This provides good evidence that the addition of chickpea starch (amylose-26% and amylopectin-74%) (Pearson & Gillett, 1999) into comminuted meat may enhance the textural properties of meat products.

2.7.7.2 Texture profile analysis (TPA)

Meat tenderness can be measured objectively, i.e., by machine/instruments. The most widely and universally used is the TPA method. Texture analysis is primarily concerned with the evaluation of mechanical characteristics where a material is subjected to a controlled force from which a deformation curve of its response is generated. These mechanical characteristics in food can be further sub-divided into primary and secondary sensory characteristics which have proven to be correlated to sensory perception (Civille & Szczesniak, 1973; Szczesniak, 1963) (Table 2.4).

This method has been extensively used with various protein gel meat products, including konjac/low-fat bologna (Chin et al., 1998), fat, starch, and egg white/bologna (Carballo et al., 1996), soy, oat bran/frankfurters (Chang & Carpenter, 1997), wheat germ protein/comminuted meat (Gnanasambandam & Zayas, 1992) and low-fat bologna (Shand, 2000).

Table 2.4 Definitions of textural characteristics[a]

Primary characteristics

Parameter	Instrumental definition	Sensorial definition
Hardness	Force necessary to attain a given deformation (Peak force of the first compression cycle).	Force required to compress a food between molars. Defined as force necessary to attain a given deformation.
Adhesiveness	The negative area for the first bite, representing the work necessary to pull compressing probe away from sample.	The work necessary to overcome the attractive forces between the surface of the food and the surface of other materials with which the food comes into contact (e.g. tongue, teeth, palate). Work required to pull food away from a surface.
Cohesiveness	Extent to which a material can be deformed before it ruptures. (The ratio of positive force during the second to that of the first compression cycle)	Degree to which a substance is compressed between the teeth before it breaks.
Springiness	Rate at which a deformed material goes back to its undefomed condition after the deforming force is removed.	Degree to which a product returns to its original shape once it has been compressed between the teeth.

Secondary characteristics

Parameter	Instrumental definition	Sensorial definition
Brittleness (Fracture force)	The first significant break in the first compression cycle.	Force at which a material fractures. Related to the primary parameters of hardness and cohesiveness, where brittle materials have low cohesiveness. Not all foods fracture and thus value may relate to hardness if only single peak is present. Brittle foods are never adhesive.
Chewiness	Energy required to masticate a solid food to a state ready for swallowing; a product of hardness, cohesiveness and springiness	Length of time required to masticate the sample, at a constant rate of force application, to reduce it to a consistency suitable for swallowing.

[a] (Modified from Civille & Szczesniak, 1973; Szczesniak, 1963)

2.7.7.3 Torsion analysis of meat batters

Another fundamental test, torsion analysis, is used on food gels for evaluating textural properties. In a torsion test, a dumbbell-shape (or capstan shape) specimen is twisted in a viscometer, with the shear stress and shear strain being measured at the fracture point. For protein gels, shear stress at failure measures gel hardness, while shear strain at failure (SA) represents the deformation (or ductility) of gelled meat products (Labudde & Lanier, 1985).

To describe textural properties of meat batters more clearly, Lanier (1986) proposed a plot of torsional rigidity vs. strain that would adequately describe the mechanical textural properties including tough, brittle, mush and rubbery (Figure 2.3). Torsional rigidity was defined as indicated in equation 2.3.

$$\text{Torsional rigidity (kPa)} = \text{Fracture stress/strain} \qquad [2.3]$$

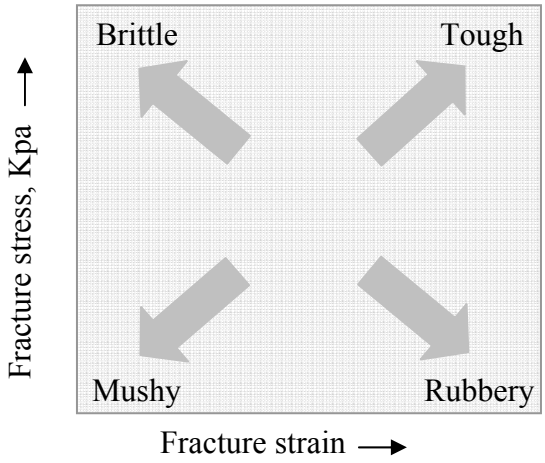

Figure 2.3 Torsion texture map (Modified from Lanier, 1986).

At high torsional rigidity and low shear strain, the mechanical texture corresponds to "brittle". Values that are high shear strain but low in torsional rigidity correspond to the mechanical texture "rubbery". High torsional rigidity and shear strain correspond to a mechanical

texture of “tough”. Conversely, low torsional rigidity and shear strain result in the texture “mushy” (Lanier, 1986).

2.7.8 Sensory evaluation

Sensory properties are among the major concerns for the utilization of plant ingredients in meat products because plant ingredients may give their indigenous flavour note to the end product. Sensory evaluation can be defined as; “A scientific discipline used to evoke, measure, analyze and interpret reactions to those characteristics of foods and materials as they are perceived by the senses of sight, smell, taste, touch and hearing” (Anonymous, 1975).

2.7.8.1 Descriptive sensory study of meat products

Basically, sensory evaluation tests fall under three types, including discrimination tests (are the products different in any way?), descriptive analysis (what are the intensities of specific attributes?), affective/hedonic tests (is the product liked?/which product is preferred?). In order to get complete knowledge about meat products with chickpea ingredients, the descriptive sensory method is a good choice. Descriptive sensory analysis involves trained or semi-trained panelists and it requires time (to set up and conduct tests) and effort to select and train the appropriate panelists for judging the samples, because panelists must be able to detect and describe the sensory attributes of a sample as well as differentiate and rate the intensity of each characteristic in the sample (Stone & Sidel, 2004). It has been widely used for analyzing sensory quality of meat products which containing plant ingredients. Serdaroğlu et al. (2005) reported that addition of legume flour (bean, chickpea, and lentil) at a level of 10%, as a meatball extender, had no effect on appearance and flavour scores. According to sensory evaluation results all meatball treatments had high overall palatability. The addition of 10% cowpea flour decreased flavour scores of chicken nuggets (Prinyawiwatkul *et. at*., 1997). Modi et al. (2003) investigated the effects of various legume flours (soybean, Bengal gram, green gram, and black

gram) on the quality of buffalo burgers, and they observed that none of the legumes had a detrimental effect on sensory properties at the level of 8% binders used.

2.7.8.2 Panelists: selection and training

There are a few basic steps that should be followed before starting descriptive analysis. Once individuals have indicated a willingness to participate, they are required to participate in a series of screening tests to determine their level of skill. The acceptance or rejection of a potential panelist is based on his or her interest, availability and health. During training for evaluating flavour and texture, each panelist is given samples of extreme cases of each attribute that will be used in the future study to some extent. Panelists gradually get familiarized with the definition of attributes (in the score sheets), number of samples and improve their sensitivity and memory for scoring flavour and texture (Cross, Moen, & Standfield, 1978). Panelists can be trained in groups, and can discuss their problems in order to clear up possible misunderstandings.

2.7.8.3 Performing sensory studies

Most product tests are fielded using individual booths. This allows panelists to assess samples without bias from other panelists. The other advantage of using a sensory laboratory is that there is more control in product preparation and presentation. The evaluation room should be clean, quiet, well ventilated and have a temperature of 22-24°C with a relative humidity of 45-55% (Meilgaard, Civille, & Carr, 1991). Ventilation in the preparation area is especially important for those products with aromatic properties. Lighting in the booth area is fluorescent. The use of various types of coloured lights (e.g. red, yellow, and blue) in the booth is recommended when necessary to mask visual differences between samples (Stone & Sidel, 2004).

3.0 EXPERIMENTAL

3.1 Study I: Studies on physico-chemical, thermal, and functional properties of seed, flour, starch and protein isolate from different Western Canadian chickpea cultivars

3.1.1 Materials

Chickpea (*Cicer arietinum* L.) samples, both Desi (*var.* CDC Cabri, CDC Vanguard and Myles) and Kabuli (*var.* CDC Xena, CDC Frontier and Amit) from the 2005 and 2006 harvests were obtained from the Crop Development Centre, College of Agriculture and Bioresources, University of Saskatchewan, Saskatoon, SK, Canada. Electrophoresis reagents were purchased from Bio-Rad Laboratories (Hercules, CA, U.S.A.). Other laboratory chemicals (analytical reagent grades) were purchased from Fisher Scientific (Pittsburgh, PA., U.S.A.) or Sigma Chemical Co. (St. Louis, MO., U.S.A.). Purified soy glycinin (11S) and vicilin (7S) were kindly provided by J. Wanasundara, Agriculture and Agri-Food Canada (AAFC), Saskatoon, SK, Canada. Samples of soy and native pea proteins were obtained from a previous study (Shand, Ya, Pietrasik, & Wanasundara, 2007) of the research group.

3.1.2 Physical properties

The physical properties of Kabuli and Desi chickpea seeds were assessed. The following physical properties were determined in duplicate (Williams, Nakoul, & Singh, 1983).

Seed weight (g/seed): recorded as the mean weight of 1000 seeds (counted twice).

$$\text{Seed weight} = \frac{\text{Weight (g)}}{\text{1000 seeds}} \qquad [3.1]$$

Seed volume (mL/seed): 150 seeds were transferred to a 250-mL graduated cylinder and 100 mL of deionized water was added. Seed volume was determined according to following equation:

$$\text{Seed volume} = \frac{\text{Total volume (mL) - 100 (mL)}}{150} \quad [3.2]$$

Hydration capacity (HC, g/seed): A fifty-gram sample of chickpea seeds was counted and transferred to a 250-mL graduated cylinder and 100 mL of deionized water was added. The cylinder was stoppered and left overnight at room temperature. The next day, the grains were blotted with absorbent paper to remove superfluous water and the swollen seeds were re-weighed. The hydration capacity per seed was determined by the following formula:

$$\text{Hydration capacity} = \frac{\text{Weight of soaked seeds (g) - weight of seeds before soaking (g)}}{\text{Number of seeds}} \quad [3.3]$$

Hydration index (HI): estimated according to following equation:

$$\text{Hydration index} = \frac{\text{Hydration capacity}}{\text{Seed weight}} \quad [3.4]$$

Swelling capacity (SC, mL/seed): A fifty-gram sample of seeds was soaked in 100 mL of de-ionized water overnight in a 250-mL graduated cylinder. The volume of the soaked seeds was noted to calculate swelling capacity per seed according to the following equation:

$$\text{Swelling capacity} = \frac{\text{Volume after soaking (mL) – Volume before soaking (mL)}}{\text{Number of seeds}} \quad [3.5]$$

Swelling index (SI): estimated as the SC/seed volume ratio.

$$\text{Swelling index} = \frac{\text{Swelling capacity}}{\text{Seed weight}} \quad [3.6]$$

Seed coat (SCT, %); 150 seeds were soaked in deionized water overnight (16 h) at room temperature (18-20°C) in order to facilitate seed coat removal. The two fractions (seed coat and cotyledon) were separated manually and the separated components were dried at 40°C in a convection oven to a constant weight. Weight of the seed coat fraction was calculated as ratio of wt of seed with intact components.

$$\text{Seed coat (\%)} = \frac{\text{Weight of seed coat (dry wt) (g)}}{\text{Weight of seeds (dry wt) (g)}} \quad [3.7]$$

3.1.3 Colour characteristics

Colour measurements of chickpea flour and protein isolates with or without defatting were carried out using a Hunterlab colourimeter D65 optical Sensor (Hunter Associates Laboratory Inc., Reston, VA, USA) on the basis of L*, a* and b* values. A glass Petri dish containing the sample (5 mm thickness) was placed above the light source, covered with a dark plate and L*, a* and b* values were recorded. The instrument (45º / 0º geometry, 10º observer) was calibrated against a standard white-coloured reference tile (L_s = 92.80 a_s = -1.25 and b_s = 0.97). Total colour difference (ΔE) was calculated by applying the following equation (Kaur et al., 2005):

$$\Delta E = [(L_s - L^*)^2 + (a_s - a^*)^2 + (b_s - b^*)^2]^{1/2} \quad [3.8]$$

Where, the L* value indicates the lightness, 0-100 representing dark to light, the a* value indicating more red. The b* value indicates the degree of the yellow-blue colour, with a higher positive b* value indicating more yellow.

3.1.4 Flour preparation

First the seeds were cleaned and any foreign materials were removed. The chickpea samples were not dehulled prior to milling for flour because preliminary studies showed separating the tightly adhered hull especially from Kabuli chickpea varieties resulted in removing a considerable amount of cotyledon materials. The milling was carried out using a turbo impact mill (Centrifugal Impact Mill Grinds, Munson Machinery Co., Inc, U.S.A.), and the resulting flour was passed through a pin mill (Cyclone sample mill, Udy Corporation, U.S.A.) with a 0.1 mm stainless steel screen. Flours were then placed in airtight containers and stored at 4°C.

3.1.5 Wet-milling process to obtain chickpea starch

Isolation of chickpea starch was carried out using whole seeds as described by Han & Tyler (2003) with modifications (Figure 3.1). Briefly, chickpea seeds were washed with deionized water and then soaked in three volumes of 0.05% (w/v) sodium bisulfite at room temperature overnight. After decanting the soaked water and washing with deionized water, the hull was removed manually by hand rubbing. Dehulled water-soaked chickpeas were milled with two volumes (w/v) of deionized water in a Waring blender at medium speed for 30 s, and then at high speed for another 30 s. The resulting slurry was then mixed with a large amount of deionized water and wet-sieved by passing through 50, 100 and finally 200 mesh screens (Tyler, Fisher Scientific Co., Pittsburg, PA, U.S.A.). The collected filtrate slurry containing starch was then centrifuged at 3000 × *g* for 10 min at 4°C. It was then vacuum filtered through two layers of Whatman *#4* filter paper, washed twice with 95% (v/v) ethanol and once with acetone, and dried at 40°C for 12 h.

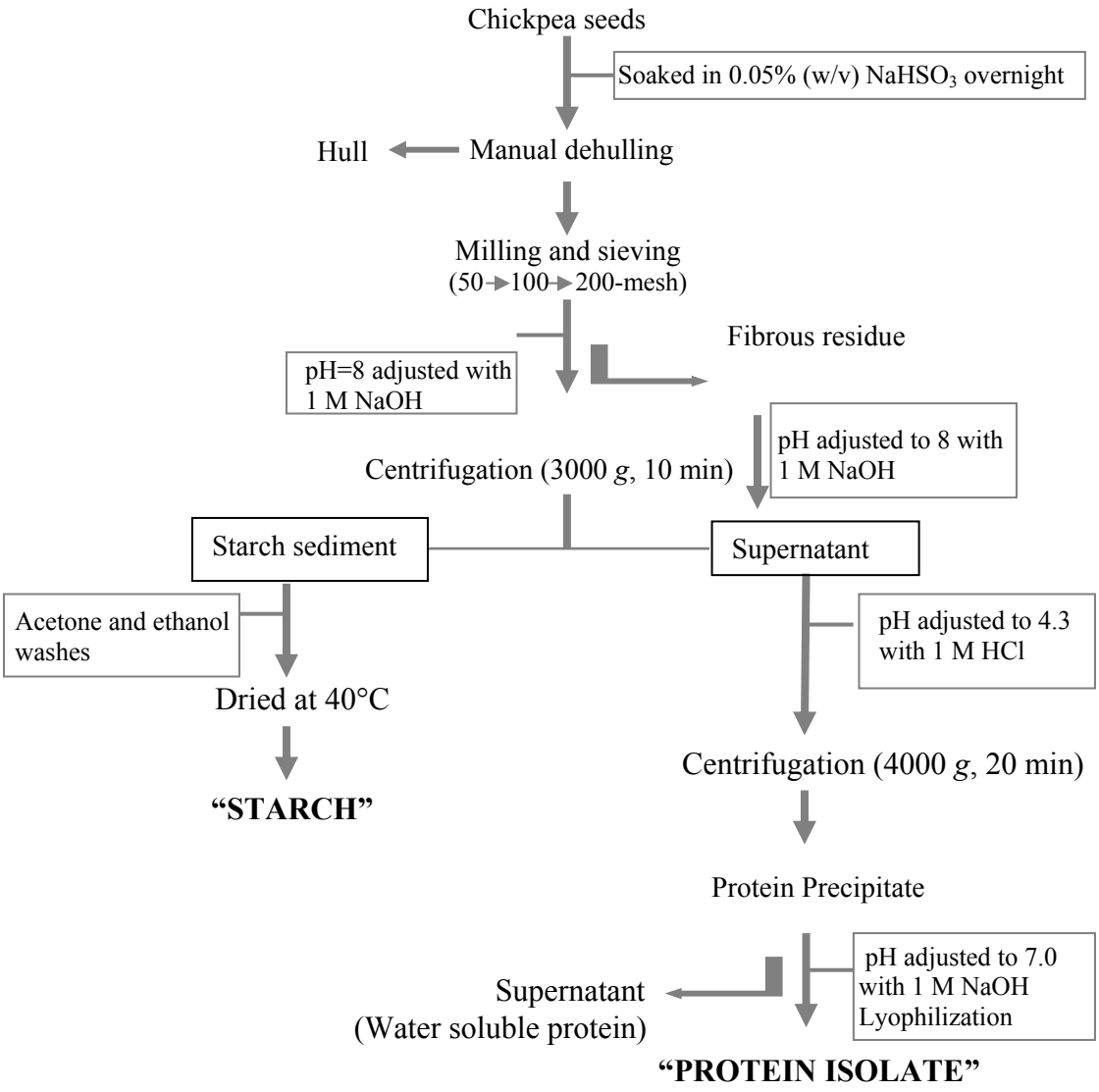

Figure 3.1 Flow-chart for isolation of chickpea proteins and starch by wet processing (Modified from Han & Tyler, 2003).

3.1.6 Separation of chickpea protein isolates

Protein isolation was carried out concurrently with starch isolation. The fibrous residues (Figure 3.1) were re-extracted two more times with the same 1 M NaOH solution (pH 8.0) under similar conditions. Then all alkali extracts and supernatant remaining from starch isolation were pooled together and pH was adjusted to 4.3 (minimum solubility point for chickpea

protein isolates) (Sánchez-Vioque et al., 1999) with 1 M HCl and maintained at 4°C for 1h. After proteins were precipitated well the mixture was centrifuged at 4000 × *g* for 20 min. The sediment was washed twice with two volumes (w/v) of deionized water. The precipitated protein was re-dispensed in deionized water and the pH was adjusted to 7.0 with 1 M NaOH prior to freeze-drying. The freeze-dried protein isolates were stored in air-tight glass containers at 4°C.

3.1.7 Composition analysis

Samples were estimated for their moisture, ash, crude fat and protein ($N \times 6.25$) content by employing the standard methods of analysis (AOAC, 1990). Total lipid content was determined by the method of Folch, Lees, & Stanley (1957) as modified for plant materials by Christie (1993). Megazyme (Megazyme International Ltd, Wicklow, Ireland) total starch analysis (*K-TSTA/05/06*), amylose/amylopectin assay (*K-AMYL/04/06*) kits and dietary fibre kit *(K-TDFR/12/05)* were used for determining the total starch, amylose and dietary fibre contents, respectively, in the both chickpea flour and isolated starch.

3.1.8 Functional properties

3.1.9 Nitrogen solubility index (NSI)

NSI was determined by the standard method 46-23 of AACC (1995), and pH vs. nitrogen solubility curves for the flour and protein isolates were determined using 30 min extraction periods at pH 2.0, 3.0, 4.0, 4.2, 4.4, 4.8, 5.0, 6.0, 7.0, 8.0, 9.0, 10.0, and 11.0. The pH levels were maintained by continuous adjustment with 1.0 N HCl or NaOH as needed.

3.1.10 Water holding capacity (WHC)

WHC for chickpea flour and protein isolates was determined according to method AACC 88-04 (AACC, 1995).

3.1.11 Oil absorption capacity (OAC)

For the determination of oil absorption of chickpea flour and protein isolates, the modified method of Lin, Humbert, & Sosulski (1974) was used. The samples (0.5 g) were mixed with 5 mL of corn oil (commercial) in preweighed centrifuge tubes. The contents were stirred for 1 min with a glass rod to disperse the sample in oil. After a holding period of 30 min, the tubes were centrifuged for 25 min at 3000 × *g*. The separated oil layer was then removed with a pipette and the tubes were inverted for 25 min to drain any remaining oil prior to reweighing. The oil absorption capacities were expressed as grams of oil bound per gram of the sample on a dry weight basis.

3.1.12 Emulsifying activity index (EAI) and emulsion stability index (ESI)

EAI and ESI of chickpea flour and protein isolates were determined by the turbidimetric method of Pearce & Kinsella (1978). A mixture of 6 mL of 0.1% protein solution (w/v, in 10 mM phosphate buffer pH 7.0) and 2 mL of corn oil was homogenized (12,000 rpm) using a Polytron PT 3100 homogenizer (Kinematica AG, Littau-Luzern, Switzerland) for 1 min. Then the emulsion (50 μL) was pipetted from the bottom of the container into 5 mL of 0.1% (w/v) sodium dodecyl sulfate (SDS) solution immediately (0 min) and 10 min after homogenization. Absorbance of the SDS dispersion of emulsion was measured at 500 nm (Hewlett Packard double beam spectrophotometer). Absorbance at 0 time was expressed as EAI of protein, and ESI was calculated by equation 3.9:

$$\text{ESI (min)} = \frac{T_0 \times t}{\Delta T} \qquad [3.9]$$

Where T_0 = the turbidity at 0 min after homogenization, ΔT = the change in turbidity between 0 and 10 min, t was time interval between 0 and 10 min.

3.1.13 Thermal properties

3.1.13.1 Least gelation concentration (LGC)

The least gelation concentration was determined by the method of Sathe, Deshpande, & Salunkhe (1982). Test tubes containing suspensions of 2, 4, 6, 8, 10, 12, 14, 18, and 20% (w/v) flour or isolates in 5 mL distilled water were heated for 1 h in a boiling water bath followed by rapid cooling under cold running water. The tubes were further cooled at 4°C for 2 h. LGC was the concentration above which the sample did not run down or slip when the test tube was inverted. Each sample was run in duplicate.

3.1.13.2 Rheological properties

3.1.13.2.1 Pasting properties of chickpea flour and starch

Pasting properties of isolated chickpea flours and starches were studied using a Rapid Visco Analyzer (Model RVA-4, Newport Scientific, Warriewood, Australia) with data analysis software (Thermocline). Viscosity profiles of samples from different chickpea cultivars were recorded using starch suspensions (6% for starches and 14% for flours on db, w/w, 25 g total weight). With constant stirring, the starch-water suspension was equilibrated at 50°C for 1 min, increased to 95°C at 6°C/min, there was then a holding phase at 95°C for 1.5 min, a cooling step from 95 to 50°C at 6°C/min and a final holding phase at 50°C for 2 min. Each sample was run in triplicate.

3.1.13.2.2 Dynamic rheological measurement of chickpea protein gelation

Heat-induced gel formation of chickpea isolates (CPI) was followed by dynamic rheological measurements using a controlled stress rheometer (AR1000, TA Instruments, New Castle, DE, U.S.A.) equipped with 40 mm parallel plate geometry for filling samples. The gap

between the two parallel plates was set at 1000 μm. Protein samples (12% protein basis) were prepared in deionized water and mixed for 30 min at 4°C. In order to remove air bubbles samples were placed in a vacuum chamber (30s) just before use. Approximately 132 μL sample solution was applied to the lower plate, and the upper plate was gently lowered under the control of a programmed procedure. The measurements were performed at a constant strain of 0.01, which was within the linear region, and at frequency of 1 Hz. A paraffin oil layer was applied to the edge of the parallel plate system to prevent the loss of moisture during the heating experiments. To induce gel formation, protein dispersions were consecutively heated from 25 to 95°C at a rate of 1°C/min, held there for 30 min and then cooled back to 25°C at a rate of 1°C/min and finally held 15 min at 25°C.

3.1.13.3 Differential scanning calorimetry

Thermal properties of chickpea protein isolates were determined using a differential scanning calorimeter (DSC) model DSC 2010 (TA Instruments, Lukens Drive, New Caste, DE, U.S.A.) equipped with thermal analysis software (TA Instrument Control). Protein isolates (200 mg each) were dispersed in an appropriate amount of water to form a slurry of 10% protein content. Protein-water slurries were well mixed and left for 30 min to reach equilibrium before analysis. The protein slurry (about 50 μL) was weighed accurately into a stainless-steel pan (large volume capsule), hermetically sealed, and scanned during a temperature increase from 25 to 140°C at a rate of 5 or 10°C/min. For flour and starch, 3.0 mg of chickpea starch or flour were directly placed in the DSC pan and 11μL deionized water was added (other steps were similar as described above). An empty pan was used as a reference. The instrument was calibrated using indium and zinc. Peak temperature and enthalpy were computed from the thermograms by using the supplied data processing software.

3.1.14 Sodium dodecyl sulfate-polyacrylamide gel electrophoresis

Molecular sizes of protein bands of chickpea flours and protein isolates were determined by sodium sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) according to the procedure of Laemmli (1970) under non-reducing condition. The SDS-PAGE was carried out on a slab gel (4% stacking gel, 12% separating gel) in an SDS-Tris-Glycine discontinuous buffer system. Protein isolate solutions (2 μg protein/μl) were prepared in non-reducing buffer solution (62.5 mM Tris-HCl, pH 6.8, 2%, w/v SDS, 10%,w/v glycerol, and 0.05%, w/v bromophenol blue). One microliter of the solution was loaded onto the gel with PhastGel sample applicators. Electrophoresis was performed at a constant current of 60 mA per gel for approximately 45 min. The gel was stained by 0.1% Coomassie brilliant blue in acetic acid/ethanol/water solution (10/30/60, v/v/v) and destained in the same solvent without Coomassie brilliant blue. The approximate molecular weights of polypeptide bands were determined by comparison to Bio-Rad molecular weight standards ranging from 6.5 to 200 kDa.

3.1.15 Statistical analysis

In study 1, the data was analysed in several ways. First, a one-way analyses of variance (ANOVA) of the 12 chickpea samples was run using the General Linear Models procedure of the SAS Institute Inc. (2004). For an evaluation of cultivar and year effects, mean values of cultivars from each year were subjected to analyses of variance. To assess biotype, cultivar was considered to be nested within biotype and analysis of variance was performed. In all cases, if the main effect was significant ($P<0.05$), mean separation was done using the Student-Newman-Kuels (SNK) procedure.

3.2 Study II: Physical, textural, and sensory properties of low fat bologna with added chickpea flour, starch, and protein isolates

3.2.1 Raw materials and ingredients

A bulk amount of Kabuli chickpea, *var.* CDC Xena, and Desi chickpea, *var.* Myles, seeds of 2007 harvest was obtained from the Crop Development Centre, College of Agriculture and Bioresources, University of Saskatchewan, Saskatoon, Canada. Chickpea flour and chickpea starches were prepared using the same methods as described in section 3.1.4 and 3.1.5, respectively, using food-grade materials at a pilot scale. Fiesta pea flour and Starbrite pea starch were provided by Parrheim Foods, Saskatoon, SK, Canada. Propulse™ pea protein isolates (Nutri-Pea Limited, Saskatoon, SK, Canada) and Supro soy protein isolates (Newly Weds Foods Co., Edmonton, AB, Canada) were used for the protein study. Native potato starch was a gift from Penford Food Ingredients Co. (Englewood, U.S.A.). Wheat flour was purchased from Robin Hood Mills (Saskatoon, SK, Canada). For each replication, fresh (1 to 2 d postmortem) pork buckeye mainly consisting of sirloin (*gluteus medius*) and loin muscle (*longissimus*) were obtained from a local slaughter plant (Country Choice Meats, Duck Lake, SK, Canada) and held at -1°C for 48 h prior to use.

3.2.2 Preparation of chickpea protein isolates

Chickpea protein isolates were prepared according to the following protocol at Agriculture and Agri-Food Canada's Food Research and Development Centre in St-Hyacinthe, QC, Canada. Chickpea samples were first frozen in liquid nitrogen (-180°C) and then serially ground (three passes) using a Comitrol Mill (Model 3600 Urschel Laboratory Inc., Valparaiso, IN, USA) equipped with 0.120" (first pass), 0.060" (second pass) and 0.030" (third pass) cutting head. 10 Kg of the ground flour was dispersed in 100 L of water at 55°C and the pH of the dispersion was adjusted and maintained at 8.5 with 2 N NaOH for 60 min. The mixture was then cooled to room temperature and the slurry was poured into a bucket centrifuge (Model

STM-2000, Western States, Hamilton, OH, U.S.A.) equipped with a 1 μm filter and centrifuged at 3200 rpm. The supernatant was collected, heated to 45°C and processed by ultrafiltration (UF) using a Romicon Ultrafiltration Unit (Romicon Inc, Woburn MA) equipped with 50 KDa molecular weight cut-off UF membrane (Romicon Kock CGT 3" HF 25-60 PM 50, Kock Membrane Systems Inc., Wilmington Massachusetts, U.S.A.). The retentate obtained was diafiltered (2X) and subsequently concentrated to a volume of 15.0 L. The pH of the retentate was then neutralized to pH 7.0 and freeze-dried using a SE-05 LYO-TECH freeze dryer (Lyo-San Inc., Lachute, QC, Canada).

3.2.3 Preparation of low-fat bologna

Low-fat pork bologna (LFPB) (6-kg batches) were prepared in triplicate for each of three separate studies of LFPB/flour, LFPB/starch and LFPB/protein isolates in the meat processing pilot plant at University of Saskatchewan under commercial processing conditions. The LFPB for each study were formulated to meet Canadian regulations for minimum total protein and meat protein contents of 11% (w/w) and 9.5% (w/w), respectively. The meat level for all studies was held at 62.6% (w/w) except control-II (79.5%, w/w) in the LFPB/protein study. The control formulation consisted of 62.6% (w/w) pork, 34.8% (w/w) ice water, 1.5% (w/w) NaCl, 0.30% (w/w) cure salt (containing 6.4% w/w sodium nitrite, Griffith Laboratories, Scarborough, ON, Canada) 0.5% (w/w) dextrose, 0.1% (w/w) sodium erythrobate (Unipac Packaging Products Ltd, Edmonton, AB, Canada), 0.25% (w/w) seasoning (Newly Weds Foods Co., Edmonton, AB, Canada). Pea ingredients (flour, starch and protein isolate) were used as a comparison, because they have a close compositional relationship with chickpea. Wheat flour, potato starch and soy protein isolates were also employed as ingredients as these are commonly used in commercial meat products. For the formulations of LFPB/flour (Kabuli, Desi, pea and wheat), water was replaced with 1:1 ratio of flour at two levels (2.5% and 5.0%). Similarly, in LFPB/starch, water was substituted (1:1) with 1.0% and 2.0% starch (Kabuli, Desi, pea and

potato). In the case of study of LFPB/protein, recipes were standardized on the basis of protein. The total crude protein content of the product was adjusted to 12.6% for the formulations with 1.5% protein level and 14.1% for the formulations with 3.0% protein level. Control I and Control II had a total meat protein level of 11.1 and 14.1%, respectively (Table 3.1).

Table 3.1 Composition of LFPB formulations manufactured using chickpea, pea and pea protein isolates.

Ingredients		Binder[c] (%)	Ice/water (%)	Meat protein (%)	Total protein[a] (%)
Control-I		-	34.80	11.07	11.07
Control-II		-	17.86	14.07	14.07
Kabuli[b]	1.5%	1.97	32.83	11.07	12.57
	3.0%	3.95	30.85	11.07	14.07
Desi[b]	1.5%	2.03	32.74	11.07	12.57
	3.0%	4.12	30.68	11.07	14.07
Pea	1.5%	1.83	32.97	11.07	12.57
	3.0%	3.66	31.14	11.07	14.07
Soy	1.5%	1.66	33.14	11.07	12.57
	3.0%	3.33	31.47	11.07	14.07

[a]Protein content (%) of meat, and plant protein isolates in the formulations
[b]Chickpea protein isolates
[c]The level is adjusted based on protein content of the ingredients to achieve 1.5 or 3.0% protein contribution to the formulation.

The pork muscles (buckeye) were ground though a 3.2 mm grinder plate (Biro Grinder, model AMFG-24, Marblehead, OH, U.S.A.), hand mixed and separated for each treatment which were prepared in random order. Meat, salt, cure and half of the ice water were mixed and chopped in a bowl chopper (35 L RMF Steel, Kansas City, MO, U.S.A.) at low speed (4,000 rpm, six blades) for a constant time of 3 bowl revolutions. The balance of dry ingredients and water were added to the chopper at intermediate speed (4000 rpm knife speed) for 12 revolutions. The bowl chopper was stopped and the lid and sides were scrapped to evenly distribute ingredients. Finally the meat batter was chopped (intermediate bowl and 8000 rpm knife speeds) for another 30 revolutions. The product temperature after chopping was approximately 11°C. Meat batter attached to the lid and the sides of the bowl were scrapped.

The lid on the chopper was closed and mixing (low bowl and 4000 rpm knife speed) was continued under vacuum (-0.8 bar) for total of 110 revolutions. End point temperature of the meat batter was around 12.5°C. The meat batter was transferred and stuffed using a Handtmann VF80 stuffer (Biberach / Riss, Germany) into 60 mm moisture proof casings and portioned to approximately 1000 grams. Then products were tensioned by hand and clipped with aluminum clips (×2) to prevent slippage.

A 250-mL plastic cup was also filled with batter using the stuffer for viscosity testing. Stuffed chubs were held for 1 h at 4°C, and then cooked in an air-agitated water bath. Chubs were cooked following a three step process; 30 min each at water temperature 60, 70°C, followed by cooking at 78°C to final internal temperature of 72°C. Internal temperature of the bologna was monitored using both an Omega digital thermometer (Omega Engineering, Inc., Stamford, CT., U.S.A) with a chromel-alumel (Omega K) thermocouple probe positioned in the geometric center of the bologna and a data logger (Barnant Scanning Thermocouple Thermometer, Burrington, III) with copper constantan thermocouples. When the endpoint temperature was achieved, the bologna were immediately chilled using cool water (20°C) to an internal temperature of 35°C (45-50 min). Bologna were stored at -1°C for 48 h before sampling for sensory and other texture analysis.

3.2.4 Chemical analysis

The moisture content, crude protein (N × 5.7 for wheat flour, × 6.25 for all other), crude fat (petroleum ether extractable) and total ash was determined in triplicate according to AOAC (1990) procedures 950.46, 976.21, and 977.14, respectively. Total starch was determined as indicated in section 3.1.7.

3.2.5 Batter viscosity

Immediately after stuffing (before thermal treatment), batter viscosity was measured in triplicate with a Brookfield Synchro-Lectric viscometer (Model RVT; Brookfield Engineering, Stoughton, MA., U.S.A.) set at 10 rpm. The number 7 spindle was positioned in a 250 mL plastic cup filled with batter and allowed to rotate for 30 s before each reading was taken. Apparent viscosity in centipoises was obtained. The temperature of each sample at the time of viscosity testing was also recorded (Shand, 2000).

3.2.6 Expressible moisture (EM)

Expressible moisture of processed bologna was determined as described by Shand (2000). Briefly, two pieces of Whatman #3 filter paper, 5.5 cm in diameter, and one piece of Whatman #50, 7.0 cm, were folded into a thimble shape to fit inside a 50-mL polycarbonate centrifuge tube. A sample of bologna (1.5±0.3 g) was placed in the thimble. The Whatman #50 paper was positioned adjacent to the meat surface and prevented the meat from sticking to the filter paper. Then the thimble containing sample was centrifuged for 15 min at 2400 × *g* (Beckman, J2-HC centrifuge with Beckman Coulter JA-17 rotor, MN, U.S.A.). The meat "cake" inside the thimble was taken out by using a pair of tweezers, and reweighed. All bologna samples were analyzed in triplicate and EM was calculated using equation 3.10.

$$\text{EM (\%)} = \frac{\text{Original sample weight (g)} - \text{final sample weight (g)}}{\text{Original sample weight (g)}} \times 100 \qquad [3.10]$$

3.2.7 Purge

Purge drip (loss) of processed bologna was monitored for sliced bologna in duplicate per treatment. Eight slices (2 stacks of 4) of bologna (3-mm thick) were vacuumed packaged (-0.9 bar, KOMET, Vacuboy, KOMET Maschinenfabfik GmbH, Plochingen, Germany) in preweighed polyethylene bags. Packages were stored in an upright position for 14 days at 4°C.

After the vacuum seal was broken, the fluid was drained and the bologna slices reweighed. Purge drip was calculated as follows:

$$\text{Purge (\%)} = \frac{\text{Initial sample weight (g) - Sample weight after 14-day drip (g)}}{\text{Initial sample weight (g)}} \times 100 \quad [3.11]$$

3.2.8 Cooking yield

Each of the raw chubs was weighed soon after processing. After bologna was cooked and chilled for 24 h, the products were reweighed. Two chubs per treatment were opened and the weight of cooked bologna was measured. Cook yield was calculated as follows (3.12):

$$\text{Cook yield (\%)} = \frac{\text{Initial bologna weight (g) - Weight after cooking (g)}}{\text{Initial bologna weight (g)}} \times 100 \quad [3.12]$$

3.2.9 Colour measurements

Colour of LFPBs was determined using a Hunterlab Miniscan XE^{TM} (Hunter Associates Laboratory, Inc., Reston, VA, U.S.A.) using illuminant A and observer 10. The instrument was standardized using black and white tiles. The colour test was performed 3 days after LFPB production (a day after sensory assessment). The samples were prepared by removing 3-mm thick slices with an electric meat slicer (SOM-12-E, Larry Sommers Limited, Toronto, ON, Canada). They were then vacuum-packed and stored 4°C for 24 h in dark. Reading were taken for L* = lightness, a* = redness, and b* = yellowness through the intact packages. The samples were then rotated 90° and readings were repeated. For each treatment colour of 2 packages was read and then averaged. Colour changes of LFPB with flours were measured weekly for a one month period. Others were measured once on day 3 after preparation.

3.2.10 Textural profile analysis

All instrumental texture profile analyses (Bourne, 1978) were done on chilled (4°C) samples. For every formulation four repeated measurements were taken for each replicate and

the mean values were reported. Samples (2.5 cm thick and 3.5 cm in diameter) were cut from the center of the bologna and axially compressed (crosshead speed 50 mm/min) twice to 50% of their original height between flat plates using a TMS-Pro Texture Press (Food Technology Corp., Sterling, VA, U.S.A.) interfaced with a computer, using the software supplied by Texture Technologies Corp. (Texture Lab Pro, version 1.13-002). Typical texture profile of LFPB is shown in Figure 3.2. The following parameters were obtained:

Hardness (N) is peak of first bite (compression),
Cohesiveness: ratio of A_2/A_1,
Adhesiveness (N s) = area A_3, Where; A_3 is area under the curve due to adhesion.
Chewiness (N mm) = hardness × cohesiveness × springiness
Where; *springiness* is the distance (% or mm) the sample recovers after the first bite.

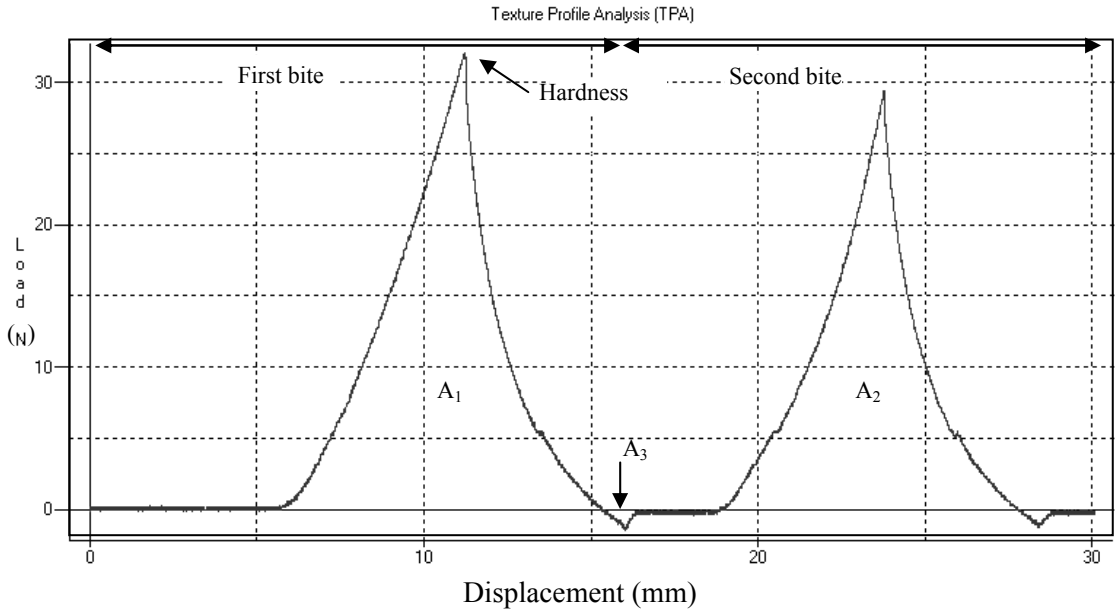

Figure 3.2 A typical texture profile analysis curve of LFPB obtained from the TMS-Pro Texture Press machine. Where, A_1, A_2 and A_3 are peak areas of relevant peaks.

3.2.11 Allo-Kramer shear

Allo-Kramer shear force (N/g) was determined by shearing a bologna square (4 × 4 cm) removed from the center of a 0.5-cm thick bologna slice. Six squares/treatment were sheared using a multi-bladed Allo-Kramer shearing device attached to TMS-Pro Texture Press

(Sterling, VA, U.S.A.). The full-scale load was set at 1000 N with the crosshead speed set at 200 mm/min. Peak shear force was recorded and divided by sample weight of each 0.5 cm square to calculate shear force in N force/g sample. Data were reported as the average of 6 readings/treatment.

3.2.12 Torsion stress and strain test

Torsion analysis was performed according to the method of Kim, Hamann, Lanier, & Wu (1986). Briefly, bologna were cut into 30-mm thick samples and a #12 cork borer was used to remove a core which was then trimmed to 28.7 mm length. Plastic disks designed to fit the torsion apparatus were glued onto the samples with cyanoacrylate glue (Loctite® 404, Instant adhesive, Loctite Corporation, Newington, CT, U.S.A.). Samples were milled onto dumbbell-shaped specimens with minimum diameter of 10.0 mm at the midsection by using a modified bench-top grinder (KCI-24A2, Bodline Electric Company, Chicago, IL, U.S.A.). The torsion apparatus was composed of a torsion fixture attached to a Brookfield digital viscometer model DV-I (Gel Consultants Inc., Raleigh, NC, U.S.A.). After placing the sample in the apparatus, the bottom plate of the torsion assembly remained stationary while the upper plate rotated at 2.5 rpm twisting the specimen until it failed. Failure shear stress and strain were calculated as described by Hamman, Saliba, & Foegeding (1989). For every treatment an average of eight samples was taken.

3.2.13 Sensory evaluation

Sensory evaluation was conducted in a room specially designed for sensory studies at the University of Saskatchewan. It is completely separated from the food preparation area and equipped with individual booths (Figure 3.3). Facing each judge seated at a booth was a small door, referred to as a "sample pass-through door". The door opens up, allowing samples to be passed to the subjects (Figure 3.3 B). This door type is known as "Bread box" type (Stone &

Sidel, 2004). The bread box is so constructed that when one side is open, the other side is closed. This minimized the likelihood of the subject (panelist) having any view of the preparation area.

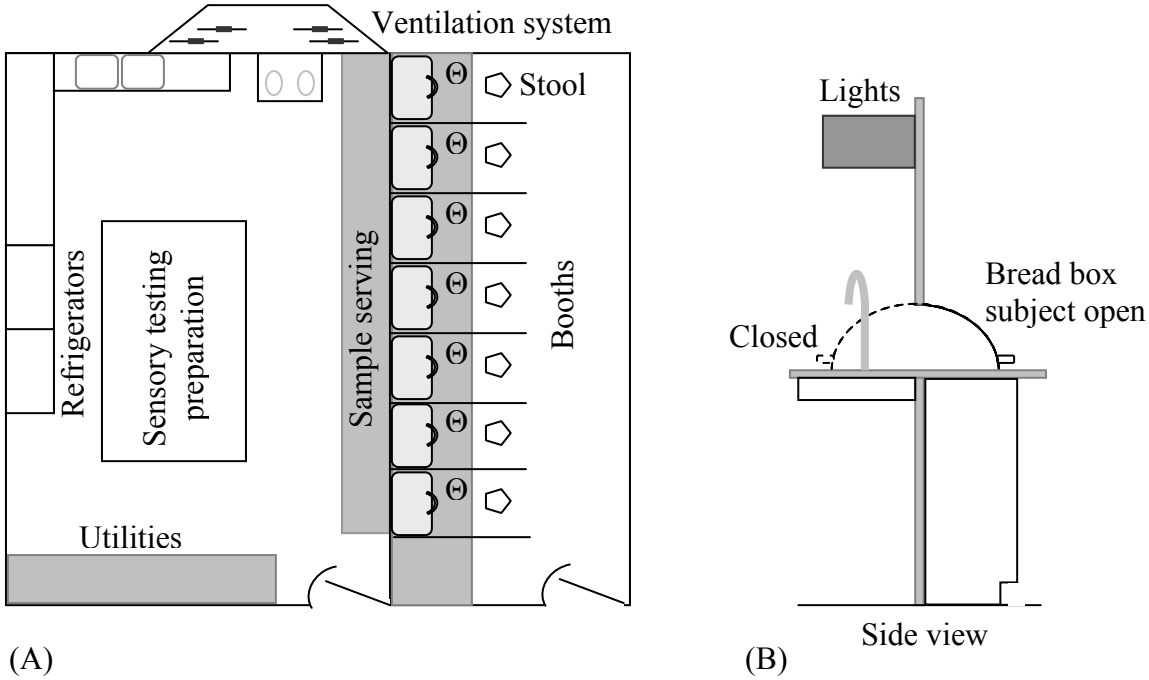

Figure 3.3 Schematic diagrams of (A) sensory test facilities showing various activities: floor plan (B) side view of product pass-through door.

Sensory evaluation was conducted by a 14-member semi-trained sensory panel using category scales. The following attributes were evaluated: Initial juiciness (1 = extremely dry, 8 = extremely juicy); firmness (1 = extremely soft, 8 = extremely firm); cohesiveness (1 = extremely brittle, 8 = extremely cohesive) overall/sustainable juiciness (1 = extremely dry, 8 = extremely juicy), saltiness (1 = not detectable, 6 = extremely salty); graininess (1 = not detectable, 6 = extremely graininess); overall flavour intensity (1 = extremely bland, 8= extremely intense); flavour desirability (1 = extremely undesirable, 8 = extremely desirable); Foreign flavour (1 = extremely intense foreign flavour, 8 = no foreign flavour); and overall acceptability (1 = extremely unacceptable, 8= extremely acceptable).

The taste panel was trained according to procedures of the American Meat Science Association (1995). Panelists were given five 30 min training sessions in which a broad range of bologna samples were presented to familiarize them with the score sheet and different flavour and texture of the products. During training each attribute was assessed by the panelist and scored and the training was continued until consensus was reached by the panelists on the rating for each attribute.

Sensory panels were carried out approximately 3 days after bologna preparation. Prior to sensory testing, samples were kept at room temperature for 15 min, and panelists were served one quarter-slices (3-mm thick) of bologna from each of eight treatments (in random order). Panelists were served with water at room temperature and salt-reduced crackers to cleanse their palate between samples. Sensory evaluation was carried out first for the flour/LFPB study, then the starch/LFPB study and finally the protein/LFPB study. Between each major study, panelists were re-trained. Samples were assessed under red lights to mask any colour difference. The proposal of this study was accepted on ethical grounds (BEH # 07-188) by the University of Saskatchewan Behavioral Research Ethics Board.

3.2.14 Statistical analysis

For texture and sensory studies, data were arranged into a randomized block design and 3 replications were conducted for all experiments (in the model, treatments and replication were included as random effects). Analysis of variance (ANOVA) using the general linear model (PROC GLM) procedure of SAS (SAS Institute Inc., 2004) was used for data analysis on means of texture and sensory data. If treatment was significant ($P < 0.05$), data were analyzed using the Student-Newman-Kuels (SNK) procedure for multiple comparisons.

4.0 RESULTS AND DISCUSSION

4.1 Study 1: Studies on physico-chemical, thermal, and functional properties of seed, flours, starches and protein isolates from Western Canadian chickpea cultivars

4.1.1 Characterization of seed and flours from some Western Canadian chickpea (*Cicer arietinum* L.) cultivars

4.1.1.1 Physical properties of chickpea seed

There are significant differences in physical characteristics among chickpea cultivars (Table 4.1.1). The seed weight of different chickpea cultivars varied significantly within each harvest year and did not vary significantly between the two harvest years. The average seed weight ranged from 0.19 to 0.47 g/seed and was significantly ($P < 0.05$) affected by biotype. This is obvious, because Kabuli-type chickpeas have large, round seeds whereas Desi-type chickpeas have small, angular seeds. CDC Xena and CDC Cabri had the highest seed weight among Kabuli and Desi-types, respectively. The average value (0.30 g/seed) was similar to the previously reported value of 0.34 g/seed (Gil et al., 1996a; Patanè et al., 2004; Williams et al., 1983). The seed volume reflected the seed weight. The highest and lowest values for seed weight corresponded to the highest and lowest values for seed volume. However, differences in seed volume between biotypes were non-significant. Seed density showed a significant ($P < 0.05$) year × cultivar interaction, but there were no significant differences among cumulative values of cultivar and biotype.

Table 4.1.1 Year × cultivar, cultivar, year and biotype effects on physico-chemical properties of chickpea varieties grown in Western Canada

Var.	Year	Bio-type[1]	Seed weight (g/seed)	Seed volume (mL/seed)	Seed density (g/mL)	Seed coat (%)
Year × cultivar						
XN	2005	K	0.46 ^{b} ± 0.02	0.36 ^{a} ± 0.01	1.26 ^{fg} ± 0.05	2.38 ^{f} ± 0.07
FT	2005	K	0.39 ^{d} ± 0.05	0.30 ^{c} ± 0.01	1.25 ^{fg} ± 0.03	1.60 ^{h} ± 0.02
AM	2005	K	0.26 ^{g} ± 0.04	0.21 ^{f} ± 0.00	1.22 ^{g} ± 0.00	1.86 ^{g} ± 0.01
CB	2005	D	0.31 ^{f} ± 0.05	0.25 ^{d} ± 0.00	1.22 ^{fg} ± 0.00	4.10 ^{b} ± 0.03
VG	2005	D	0.24 ^{i} ± 0.00	0.19 ^{g} ± 0.00	1.28 ^{ef} ± 0.00	6.44 ^{a} ± 0.10
ML	2005	D	0.19 ^{k} ± 0.01	0.15 ^{h} ± 0.00	1.26 ^{fg} ± 0.00	2.69 ^{e} ± 0.30
XN	2006	K	0.48 ^{a} ± 0.00	0.32 ^{b} ± 0.01	1.46 ^{a} ± 0.02	2.42 ^{f} ± 0.03
FT	2006	K	0.35 ^{c} ± 0.06	0.26 ^{d} ± 0.00	1.33 ^{cd} ± 0.01	1.52 ^{h} ± 0.00
AM	2006	K	0.25 ^{h} ± 0.02	0.18 ^{g} ± 0.00	1.39 ^{b} ± 0.00	1.73 ^{gh} ± 0.02
CB	2006	D	0.32 ^{e} ± 0.01	0.24 ^{e} ± 0.01	1.32 ^{ed} ± 0.06	3.69 ^{c} ± 0.04
VG	2006	D	0.22 ^{j} ± 0.06	0.16 ^{h} ± 0.01	1.37 ^{bcd} ± 0.05	2.94 ^{d} ± 0.04
ML	2006	D	0.18 ^{l} ± 0.00	0.13 ^{i} ± 0.01	1.38 ^{bc} ± 0.04	2.27 ^{f} ± 0.37
Cultivar						
XN	05/06	K	0.47 ^{a} ± 0.01	0.34 ^{a} ± 0.02	1.36 ^{a} ± 0.11	2.40 ^{ab} ± 0.05
FT	05/06	K	0.36 ^{b} ± 0.01	0.28 ^{ab} ± 0.03	1.29 ^{a} ± 0.01	1.56 ^{b} ± 0.05
AM	05/06	K	0.25 ^{d} ± 0.02	0.20 ^{bc} ± 0.01	1.31 ^{a} ± 0.00	1.79 ^{b} ± 0.07
CB	05/06	D	0.31 ^{c} ± 0.01	0.25 ^{abc} ± 0.01	1.27 ^{a} ± 0.06	3.90 ^{ab} ± 0.23
VG	05/06	D	0.23 ^{d} ± 0.01	0.27 ^{abc} ± 0.02	1.33 ^{a} ± 0.05	4.69 ^{a} ± 1.92
ML	05/06	D	0.19 ^{e} ± 0.01	0.14 ^{c} ± 0.01	1.32 ^{a} ± 0.04	2.48 ^{ab} ± 0.37
Biotype						
		Kabuli	0.36 ^{a} ± 0.09	0.27 ^{a} ± 0.07	1.31 ^{a} ± 0.09	1.92 ^{b} ± 0.37
		Desi	0.24 ^{b} ± 0.06	0.22 ^{a} ± 0.05	1.20 ^{a} ± 0.06	3.69 ^{a} ± 1.42
Year						
		2005	0.31 ^{a} ± 0.10	0.28 ^{a} ± 0.07	1.38 ^{a} ± 0.05	3.18 ^{a} ± 0.76
		2006	0.30 ^{a} ± 0.09	0.22 ^{a} ± 0.07	1.14 ^{b} ± 0.04	2.43 ^{a} ± 1.72
Average ± SD			0.30 ± 0.10	0.25 ± 0.08	1.26 ± 0.21	2.80 ± 1.40
CV (%)			32.3	33.4	16.3	49.8

Means (±SD) of triplicate analysis per year
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
[1]Biotype: K = Kabuli, D = Desi. *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles
SD = standard deviation; CV = coefficient of variation

Table 4.1.1 Continued …

Var.	Year	Geno -type	HC (g/seed)	HI	SC (mL/seed)	SI
Year × cultivar						
XN	2005	K	0.52 [a]± 0.01	1.14 [a]± 0.01	0.52 [b]± 0.03	1.43 [b]± 0.10
FT	2005	K	0.40 [c]± 0.01	1.05 [c]± 0.02	0.39 [c]± 0.00	1.28 [c]± 0.03
AM	2005	K	0.28 [g]± 0.00	1.09 [b]± 0.00	0.26 [g]± 0.00	1.20 [de]± 0.00
CB	2005	D	0.32 [e]± 0.00	1.03 [ed]± 0.01	0.30 [e]± 0.01	1.19 [de]± 0.03
VG	2005	D	0.25 [h]± 0.00	1.05 [c]± 0.00	0.25 [g]± 0.01	1.30 [cd]± 0.04
ML	2005	D	0.20 [j]± 0.00	1.02 [d]± 0.00	0.19 [i]± 0.01	1.26 [cd]± 0.04
XN	2006	K	0.51 [b]± 0.00	1.06 [c]± 0.01	0.53 [a]± 0.01	1.63 [a]± 0.02
FT	2006	K	0.38 [d]± 0.01	0.98 [f]± 0.02	0.32 [d]± 0.00	1.25 [cde]± 0.01
AM	2006	K	0.25 [h]± 0.00	1.00 [ef]± 0.00	0.25 [g]± 0.00	1.39 [b]± 0.00
CB	2006	D	0.29 [f]± 0.01	0.92 [h]± 0.02	0.28 [f]± 0.01	1.17 [e]± 0.08
VG	2006	D	0.21 [i]± 0.00	0.96 [g]± 0.00	0.21 [h]± 0.00	1.29 [c]± 0.07
ML	2006	D	0.14 [k]± 0.01	0.78 [i]± 0.03	0.13 [j]± 0.01	1.03 [f]± 0.01
Cultivar						
XN	05/06	K	0.51 [a]± 0.01	1.10 [a]± 0.04	0.53 [a]± 0.02	1.53 [a]± 0.13
FT	05/06	K	0.37 [ab]± 0.03	1.02 [a]± 0.04	0.36 [ab]± 0.04	1.27 [ab]± 0.03
AM	05/06	K	0.27 [bc]± 0.02	1.05 [a]± 0.05	0.25 [bc]± 0.00	1.30 [ab]± 0.10
CB	05/06	D	0.30 [bc]± 0.01	0.97 [a]± 0.06	0.29 [bc]± 0.01	1.18 [b]± 0.06
VG	05/06	D	0.35 [abc]± 0.02	0.97 [a]± 0.05	0.34 [abc]± 0.02	1.30 [ab]± 0.05
ML	05/06	D	0.19 [c]± 0.03	0.90 [a]± 0.13	0.16 [c]± 0.03	1.14 [b]± 0.13
Biotype						
		Kabuli	0.38 [a]± 0.11	1.06 [a]± 0.06	0.38 [a]± 0.12	1.37 [a]± 0.15
		Desi	0.28 [b]± 0.06	1.13 [a]± 0.09	0.27 [b]± 0.06	1.20 [a]± 0.11
Year						
		2005	0.37 [a]± 0.12	1.23 [a]± 0.09	0.36 [a]± 0.13	1.29 [a]± 0.20
		2006	0.29 [a]± 0.11	0.95 [a]± 0.05	0.29 [a]± 0.11	1.28 [a]± 0.09
Average ± SD			0.33 ± 0.13	1.09 ± 0.32	0.32 ± 0.13	1.29 ± 0.15
CV (%)			38.80	29.01	40.94	11.80

Means (±SD) of triplicate analysis per year
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
HC = Hydration capacity, HI = Hydration index, SC = Swelling capacity, SI = Swelling index
[1]Biotype: K = Kabuli, D = Desi. *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

The average value for seed density of 1.26 g/mL was close to the value of 1.18 g/mL reported by Patanè, Iacoponi, & Raccuia (2004) but higher than the values (0.79 – 0.85 g/mL) reported by Jood, Bishnoi, & Sharma (1998). As expected, Kabuli chickpea had less seed coat than did the Desi-type ($P< 0.05$), which was not affected by the production year. Furthermore, it was observed that Kabuli chickpea had a very thin and somewhat transparent seed coat and Desi had a thick and opaque seed coat. The mean value of 2.80% for seed coat percentage was lower than the earlier value of 4.47% reported by Patanè et al. (2004).

Water absorption and swelling properties of chickpea seed were also investigated (Table 4.1.1). Significant differences were observed in HC, HI, SC and SI for the year × cultivar interaction. Among different chickpea cultivars, the highest hydration capacity (HC), hydration index (HI), swelling capacity (SC) and swelling index (SI) were observed for CDC Xena (Kabuli-type). On the other hand, the lowest HC, HI and SC were observed for the Myles cultivar (Desi-type) for both production years. These differences among chickpea samples may be attributed to the differences in affinity and permeability of the seed coat, the physical hardness of the seed, the chemical composition of the cell wall, etc., between Kabuli-type and Desi-type chickpea seeds (Muller, 1967).

Comparison of mean values of hydration and swelling parameters of chickpea seed across years showed minor level significant differences among cultivars, with the exception of HI, which did not differ among cultivars. HC and SC were significantly lower in Desi biotype (0.28 g/seed and 0.27 mL/seed, respectively) compared to Kabuli-type (0.37 g/seed and 0.37 mL/seed, respectively). Higher water absorption of Kabuli may be attributed to the greater permeability of its thin seed coat and softer cotyledons (Muller, 1967). The mean values for hydration capacity and swelling capacity (0.33 g/seed and 0.32 mL/seed, respectively) did not differ much from that reported in the literature (Gil et al., 1996a; Patanè et al., 2004; Williams et al., 1983). However, Patanè et al. (2004) reported much lower HC and SC values for both

Kabuli and Desi-types (Kabuli = 0.26 g/seed and 0.10 mL/seed; Desi = 0.24 g/seed and 0.10 mL/seed, respectively) from some Sicilian (Italian) populations of chickpea.

Correlation studies of physico-chemical properties for different chickpea cultivars are shown in Table 4.1.2 and Figure 4.1.1. A strong positive correlation of seed weight with hydration capacity (r = 0.80, P<0.01) and swelling capacity (r = 0.80, P<0.01) was observed (Figure 4.1.1). Seed volume had a highly significant positive correlation with swelling capacity (r = 0.95, P<0.001) and hydration capacity (r = 0.97, P<0.001) (Figure 4.1.1). Similar correlations between seed volume and hydration capacity were reported by Kaur et al. (2005) for Indian chickpea varieties.

Table 4.1.2 Correlation coefficients (r) among physico-chemical properties of chickpea varieties (Combined data, n=12)

	2	3	4	5	6	7	8
1. Seed weight	0.75**	0.20	0.80**	-0.01	0.80**	0.68*	-0.21
2. Seed volume	1	-0.48	0.97***	0.63*	0.95***	0.55	0.36
3. Seed density		1	-0.39	-0.93***	-0.33	0.12	-0.80**
4. HC			1	0.58*	0.99***	0.69*	0.28
5. HI				1	0.55	0.22	0.76**
6. SC					1	0.73**	0.25
7. SI						1	-0.09
8. Seed coat							1

*, **, *** = Significant at P<0.05, 0.01 and 0.001, respectively.
HC = Hydration capacity, HI = Hydration index, SC = Swelling capacity, SI = Swelling index

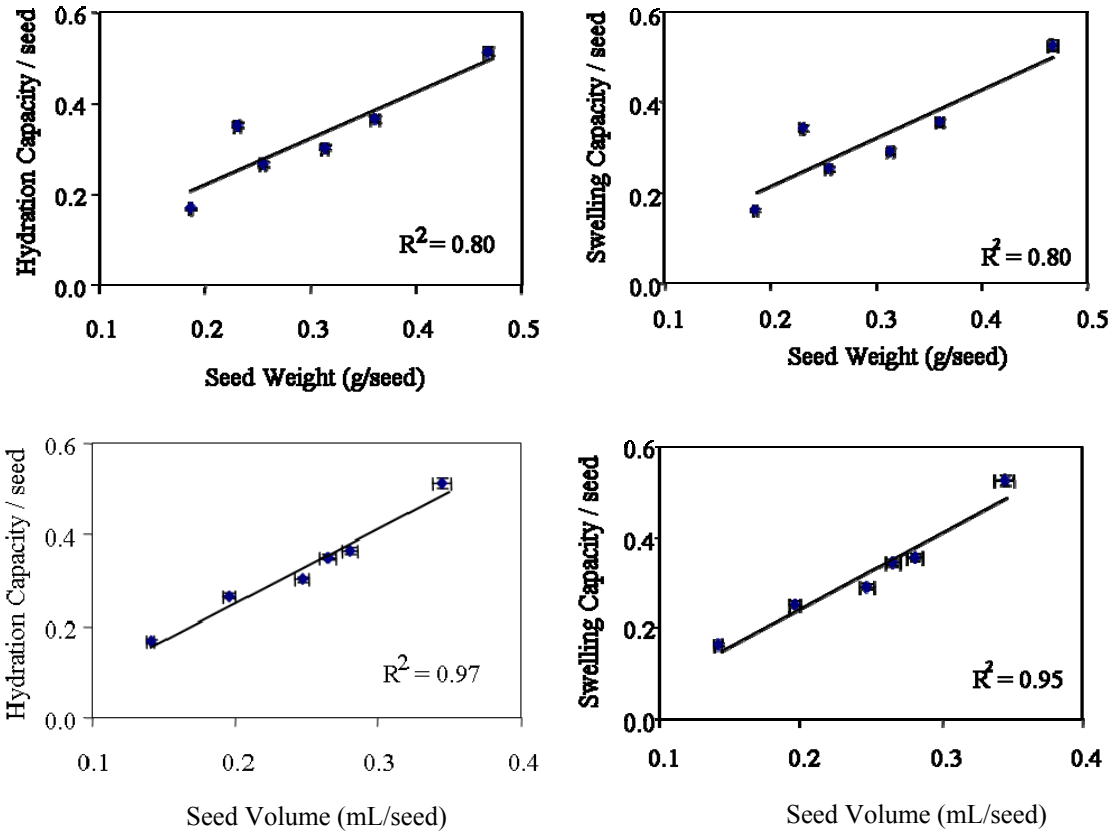

Figure 4.1.1 Relationship between hydration capacity and swelling capacity with seed weight and seed volume of Western Canadian chickpea cultivars

4.1.1.2 Colour characteristics of the flour and seeds

Seed colour is a major characteristic that usually is used to differentiate chickpea bio-types. Flour was made from various chickpeas with seed coat included. Defatted flours were also prepared. CIE colour values (L*, a*, b* and ΔE) of native flours from different chickpea cultivars are shown in Table 4.1.3. CIE (L*, a*, b* and ΔE) values of normal and defatted flour were also compared with seed colour of the corresponding varieties (Figure 4.1.2). Varietal differences were observed for various CIE colour parameters. Confirming visual appearance, Kabuli chickpea seeds had significantly higher L* values than the lightness values of Desi-type. The seed coat imparted its colour to the native flour as well, because a similar biotype effect for L* value can be seen by native flours. The high L* parameter and ΔE for all flours indicates

that they are a light colour in nature. But L* values for seed were lower than L* values of native flours. The a* value, an indicator of redness, of Desi seeds was higher ($P < 0.05$) than that of Kabuli-type. Flours from all varieties exhibited low a* values, which indicate a light green colour of these samples. However, chickpea seed showed relatively higher red colour than did flours. Lightness and redness values of flours from the 2005 harvest were significantly different from flour from the 2006 harvest. The b* value, an indicator of (-) blue and yellow (+), did not show either biotype or cultivar differences for chickpea flour. However, chickpea seeds from Kabuli-types had more yellow colour than that of the seed from Desi-type (Figure 4.1.2).

During the total lipid analysis (in proximate analysis), the extracted chickpea lipid was a brilliantly clear oil with a light golden yellow colour. The colour may be due to naturally occurring carotenoid pigments. Usually, these fat soluble pigments gave the yellowness to the flours. Defatted flour showed higher L* values when compared to the normal, fat-containing flours (Figure 4.1.2). Both native and defatted flours from the 2006 harvest were lighter (L* and ΔE) ($P < 0.05$) than the samples obtained from the 2005 harvest. This may be due to the shorter storage time following harvest of 2006 samples (9 months for 2005 and 3 months for 2006 cultivars).

According to the results (Figure 4.1.2), defatted flours showed much lower b* values. Therefore, removal of fat from the native chickpea flour can be used to produce flour with off-white colour (high L* and low a*/b* values), which will be useful in food applications.

Table 4.1.3 Year × cultivar, cultivar and year effects on CIE colour values of flours from different chickpea cultivars

Var.	Year	Bio-type [1]	L*	a*	b*	ΔE
Year × cultivar						
XN	2005	K	86.85 ^{cd} ± 0.17	1.64 ^{g} ± 0.06	20.30 ^{b} ± 0.18	32.93 ^{e} ± 0.36
FT	2005	K	87.13 ^{bc} ± 0.18	1.71 ^{g} ± 0.05	20.50 ^{b} ± 0.22	34.03 ^{f} ± 0.28
AM	2005	K	86.53 ^{d} ± 0.07	1.89 ^{ef} ± 0.10	20.81 ^{ab} ± 0.07	33.81 ^{ef} ± 0.23
CB	2005	D	81.00 ^{g} ± 0.44	1.93 ^{e} ± 0.10	19.04 ^{c} ± 0.37	19.30 ^{a} ± 1.30
VG	2005	D	81.01 ^{g} ± 0.32	1.89 ^{ef} ± 0.07	19.28 ^{c} ± 0.12	19.72 ^{ab} ± 0.28
ML	2005	D	82.56 ^{e} ± 0.36	1.77 ^{fg} ± 0.06	18.33 ^{d} ± 0.19	20.67 ^{b} ± 0.27
XN	2006	K	88.14 ^{a} ± 0.15	1.70 ^{g} ± 0.06	18.90 ^{c} ± 0.31	32.83 ^{e} ± 0.66
FT	2006	K	87.36 ^{b} ± 0.17	2.22 ^{d} ± 0.06	21.10 ^{a} ± 0.18	36.71 ^{g} ± 0.36
AM	2006	K	87.48 ^{b} ± 0.08	2.28 ^{d} ± 0.01	20.79 ^{ab} ± 0.04	36.48 ^{g} ± 0.09
CB	2006	D	82.41 ^{e} ± 0.20	2.52 ^{c} ± 0.03	20.35 ^{b} ± 0.13	25.91 ^{c} ± 0.72
VG	2006	D	81.63 ^{f} ± 0.25	3.09 ^{a} ± 0.07	20.71 ^{ab} ± 0.08	26.23 ^{c} ± 0.25
ML	2006	D	82.68 ^{e} ± 0.04	2.79 ^{b} ± 0.03	20.41 ^{b} ± 0.04	27.12 ^{d} ± 0.12
Cultivar						
XN	05/06	K	87.50 ^{a} ± 0.73	1.67 ^{b} ± 0.06	19.60 ^{a} ± 0.82	32.88 ^{a} ± 0.46
FT	05/06	K	87.25 ^{a} ± 0.20	1.97 ^{ab} ± 0.28	20.81 ^{a} ± 0.38	35.37 ^{a} ± 1.50
AM	05/06	K	87.00 ^{a} ± 0.52	2.09 ^{ab} ± 0.23	20.80 ^{a} ± 0.05	35.15 ^{a} ± 1.47
CB	05/06	D	81.71 ^{bc} ± 0.83	2.23 ^{ab} ± 0.33	19.70 ^{a} ± 0.76	22.61 ^{b} ± 3.74
VG	05/06	D	81.32 ^{c} ± 0.43	2.49 ^{a} ± 0.66	20.00 ^{a} ± 0.79	22.98 ^{b} ± 3.58
ML	05/06	D	82.62 ^{b} ± 0.24	2.28 ^{ab} ± 0.56	19.37 ^{a} ± 1.15	23.90 ^{b} ± 3.54
Biotype						
		Kabuli	87.24 ^{a} ± 0.54	1.91 ^{a} ± 0.27	20.40 ^{a} ± 0.76	34.47 ^{a} ± 1.64
		Desi	81.88 ^{b} ± 0.76	2.33 ^{a} ± 0.52	19.69 ^{a} ± 0.90	23.16 ^{b} ± 3.45
Year						
		2005	84.18 ^{b} ± 2.80	1.80 ^{b} ± 0.46	19.71 ^{a} ± 0.93	26.74 ^{b} ± 7.08
		2006	84.95 ^{a} ± 2.82	2.43 ^{a} ± 0.12	20.38 ^{a} ± 0.74	30.88 ^{a} ± 4.80

Means (±SD)of triplicate analysis per year
Means followed by same letter within a column and section do not differ significantly ($P < 0.025$)
[1] Biotypes: K = Kabuli, D = Desi . *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles.
CIE colour: L* = lightness; a* = redness; b* = yellowness

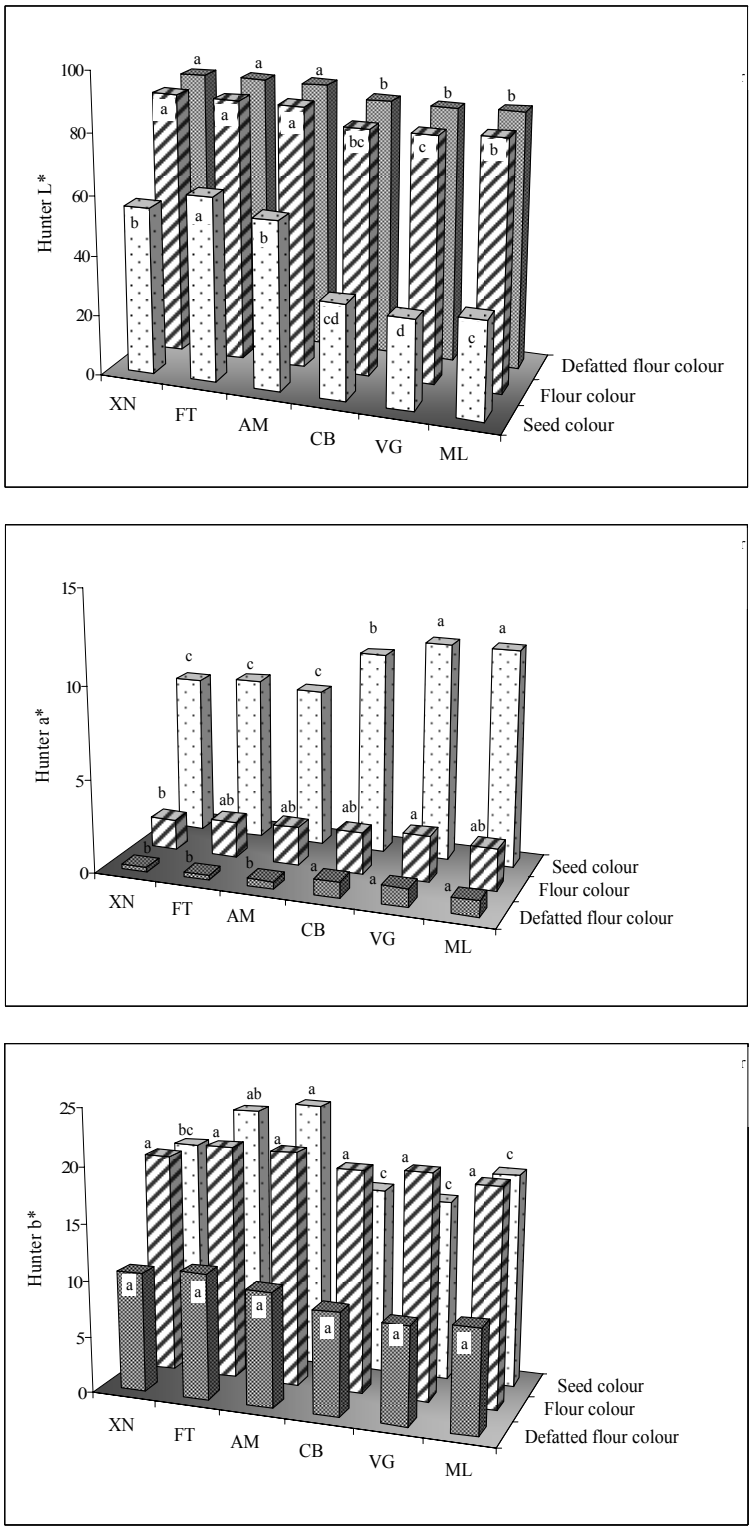

Figure 4.1.2 CIE colour values for seed, flour and defatted flour of six Western Canadian chickpea varieties from 2005 and 2006 harvests. [a-c] Means within CIE values of seed, flour and defatted flour with same latter are not significantly different (P > 0.05). CIE colour: L* = lightness; a* = redness; b* = yellowness. *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles.

4.1.1.3 Proximate composition of chickpea flour

The proximate composition of chickpea flour varied significantly among different chickpea cultivars (Table 4.1.3) when compared within each year. Reflecting the biotype effect ($P<0.05$), Kabuli chickpeas had higher protein content (by approximately 1.5%) than those of the Desi-type. Among different Western Canadian chickpea cultivars, protein contents ranged from 22.8 to 24.9% for Kabuli and from 21.8 to 23.5% for Desi biotypes. However, ash, crude fat and total fat analysis did not show any significant differences between Kabuli and Desi biotypes. The ash, crude fat and total fat contents of cultivars ranged from 2.8-3.0%, 6.7-7.6%, and 7.3-8.0%, respectively. Due to extraction of both polar and non-polar lipids, total fat content of each cultivar was slightly higher than the crude fat of the corresponding cultivar.

There were no differences between years for ash and total fat ($P<0.05$). Mean values for protein, carbohydrate, lipid, and ash content of 22.5, 69.5, 5.01 and 2.98%, respectively, for chickpea flour have been reported earlier (Milan-Carrillo et al., 2000), which were similar to the protein and ash contents in the present study.

Since the amylose content of starch plays a very important role in the gelatinization and pasting properties of flour, amylose content was determined on defatted flour. Biotypes showed significantly different amylose contents but differences were small. Amylose contents of different chickpea cultivars ranged from 17.7 - 23.1% for Kabuli-type and 21.2 - 23.2% for Desi-type (Table 4.1.5). There were biotype and year effects for total starch content. In a cultivar comparison, Kabuli-type chickpea had higher total starch content (45.3 - 49.8%) than did Desi-type (41.1 - 43.1%). Meares et al. (2004) reported that Desi and Kabuli chickpea cultivars had amylose content of 26.1 and 26.4%, and total starch content of 45.2 and 42.1%, respectively.

Table 4.1.4 Year × cultivar, cultivar, year and biotype effects on chemical composition of chickpea flour on a dry weight basis

Var.	Year	Geno -type[1]	Protein[2] (%)	Ash (%)	Crude fat (%)	Total fat (%)
Year × cultivar						
XN	2005	K	25.2 ^{a} ± 0.02	3.0 ^{b} ± 0.03	6.7 ^{d} ±0.03	7.8 ^{bced} ± 0.04
FT	2005	K	22.3 ^{e} ± 0.09	2.8 ^{de} ± 0.04	7.1 ^{bc} ±0.01	7.0 ^{h} ± 0.05
AM	2005	K	23.9 ^{bc} ± 0.01	2.9 ^{c} ± 0.02	6.9 ^{c} ±0.00	7.6 ^{defg} ± 0.00
CB	2005	D	21.2 ^{f} ± 0.04	2.7 ^{e} ± 0.03	7.3 ^{b} ±0.05	8.1 ^{bc} ± 0.01
VG	2005	D	21.7 ^{f} ± 0.02	2.7 ^{e} ± 0.02	7.6 ^{a} ±0.04	8.2 ^{b} ± 0.16
ML	2005	D	23.9 ^{bc} ± 0.08	2.9 ^{c} ± 0.02	6.5 ^{e} ±0.01	8.7 ^{a} ± 0.14
XN	2006	K	24.5 ^{b} ± 0.14	2.9 ^{c} ± 0.06	6.9 ^{c} ±0.04	7.2 ^{gh} ± 0.11
FT	2006	K	23.3 ^{c} ± 0.02	3.1 ^{b} ± 0.08	7.2 ^{b} ±0.01	7.6 ^{defg} ± 0.04
AM	2006	K	24.1 ^{b} ± 0.17	3.0 ^{b} ± 0.00	7.5 ^{a} ±0.17	8.0 ^{bcd} ± 0.04
CB	2006	D	22.4 ^{e} ± 0.85	3.0 ^{b} ± 0.01	7.6 ^{a} ±0.02	7.4 ^{efgh} ± 0.25
VG	2006	D	22.6 ^{de} ± 0.12	2.8 ^{cd} ± 0.05	7.6 ^{a} ±0.03	7.7 ^{cdef} ± 0.04
ML	2006	D	23.1 ^{cd} ± 0.02	3.1 ^{a} ± 0.02	6.9 ^{c} ±0.12	7.3 ^{fgh} ± 0.09
Cultivar						
XN	05/06	K	24.9 ^{a} ± 0.51	2.9 ^{a} ± 0.09	6.8 ^{b} ± 0.18	7.5 ^{a} ± 0.44
FT	05/06	K	22.8 ^{bcd} ± 0.70	2.9 ^{a} ± 0.21	7.2 ^{ab} ± 0.09	7.3 ^{a} ± 0.43
AM	05/06	K	24.0 ^{ab} ± 0.16	3.0 ^{a} ± 0.12	7.2 ^{ab} ± 0.41	7.8 ^{a} ± 0.27
CB	05/06	D	21.8 ^{d} ± 0.83	2.8 ^{a} ± 0.24	7.4 ^{a} ± 0.23	7.8 ^{a} ± 0.53
VG	05/06	D	22.1 ^{cd} ± 0.61	2.8 ^{a} ± 0.11	7.6 ^{a} ± 0.01	8.0 ^{a} ± 0.35
ML	05/06	D	23.5 ^{abc} ±0.54	3.0 ^{a} ± 0.16	6.7 ^{b} ± 0.31	8.0 ^{a} ± 1.01
Biotype						
		Kabuli	23.9 ^{a} ± 1.00	2.9 ^{a} ± 0.12	7.0 ^{a} ± 0.39	7.5 ^{a} ± 0.57
		Desi	22.5 ^{b} ± 0.96	2.9 ^{a} ± 0.19	7.2 ^{a} ± 0.31	8.0 ^{a} ± 0.30
Year						
		2005	23.3 ^{a} ± 1.54	2.8 ^{a} ± 0.14	7.0 ^{a} ± 0.39	7.9 ^{a} ± 0.57
		2006	23.0 ^{a} ± 0.84	3.0 ^{a} ± 0.12	7.3 ^{a} ± 0.31	7.5 ^{a} ± 0.30
Average ± SD			23.2 ± 1.19	2.9 ± 0.15	7.1 ± 0.37	7.7 ± 4.55
CV (%)			5.1	5.3	5.2	6.2

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
[1] Biotypes: K = Kabuli, D = Desi. [2] Total nitrogen × 6.25.
Var: XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles
SD = standard deviation; CV = coefficient of variation

Table 4.1.5 Year × cultivar, cultivar, year and biotype effects on amylose, total starch and dietary fiber of chickpea flour

Var	Year	Bio-type[1]	Amylose (%)	Total starch (%)	Dietary fiber [2] (%)		
					IDF	SDF	TDF
Year × cultivar							
XN	2005	K	23.0 ^{bc}±0.34	46.5 ^{d} ±0.69	15.1 ^{e} ±0.11	7.2 ^{d} ±0.02	22.3 ^{d} ±0.10
FT	2005	K	17.6 ^{g} ± 0.22	48.9 ^{c} ±0.47	15.0 ^{e} ±0.03	7.2 ^{d} ±0.03	22.1 ^{d} ±0.06
AM	2005	K	20.0 ^{ef} ± 0.10	42.5 ^{f} ±0.45	15.9 ^{d} ±0.04	7.4 ^{c} ±0.13	23.4 ^{c} ±0.15
CB	2005	D	21.0 ^{def} ±0.05	41.0 ^{g} ±0.28	18.6 ^{c} ±0.10	7.8 ^{b} ±0.02	26.4 ^{b} ±0.12
VG	2005	D	21.4 ^{cdef} ±1.02	38.6 ^{h} ±0.62	19.5 ^{b} ±0.11	7.9 ^{a} ±0.04	27.5 ^{a} ±0.14
ML	2005	D	21.9 ^{bcd} ±0.34	37.4 ^{I} ±0.41	20.1 ^{a} ±0.17	7.5 ^{c} ±0.04	27.6 ^{a} ±0.17
XN	2006	K	23.3 ^{b} ±0.14	52.0 ^{a} ±0.38	15.7 ^{d} ±0.08	7.8 ^{ab} ±0.08	23.5 ^{c} ±0.02
FT	2006	K	17.7 ^{g} ±1.20	50.6 ^{b} ±0.49	13.9 ^{e} ±0.03	7.0 ^{d} ± 0.08	20.8 ^{d} ±0.11
AM	2006	K	19.8 ^{f} ±1.20	48.0 ^{c} ±0.49	16.2 ^{c} ±0.08	7.5 ^{bc} ±0.05	23.7 ^{c} ±0.13
CB	2006	D	21.3 ^{cdef} ±0.38	45.3 ^{ed} ±0.01	18.4 ^{b} ±0.02	7.1 ^{cd} ±0.04	25.6 ^{b} ±0.04
VG	2006	D	21.7 ^{bcde} ±0.57	45.8 ^{ed} ±0.85	19.7 ^{a} ±0.50	8.1 ^{a} ±0 .38	27.8 ^{a} ± 0.77
ML	2006	D	24.4 ^{a} ±0.46	44.7 ^{e} ±0.07	18.2 ^{b} ±0.35	7.2 ^{cd} ± 0.29	25.4 ^{b} ±0.45
Cultivar							
XN	05/06	K	23.1 ^{a} ± 0.21	49.3 ^{a} ± 3.85	15.4 ^{b} ±0.39	7.5 ^{ab} ±0.44	22.9 ^{b} ±0.82
FT	05/06	K	17.7 ^{c} ± 0.09	49.8 ^{a} ± 1.21	14.4 ^{b} ±0.78	7.1 ^{b} ±0.14	21.5 ^{b} ±0.92
AM	05/06	K	19.9 ^{b} ± 0.13	45.3 ^{b} ±3.87	16.1 ^{b} ±0.15	7.5 ^{ab} ±0.06	23.5 ^{b} ±0.21
CB	05/06	D	21.2 ^{b} ±0.18	43.1 ^{bc} ±3.05	18.5 ^{a} ±0.10	7.5 ^{ab} ±0.48	26.0 ^{a} ±0.58
VG	05/06	D	21.5 ^{ab} ±0.16	42.2 ^{bc} ±5.03	19.6 ^{a} ±0.10	8.0 ^{a} ±0.13	27.6 ^{a} ±0.23
ML	05/06	D	23.2 ^{a} ± 1.80	41.1 ^{c} ±5.15	19.2 ^{a} ±1.35	7.4 ^{ab} ±0.21	26.5 ^{a} ±1.56
Biotype							
		Kabuli	20.2 ^{b} ± 2.43	48.1 ^{a} ±3.33	15.3 ^{b} ±0.84	7.3 ^{a} ±0.29	22.6 ^{b} ±1.08
		Desi	22.0 ^{a} ± 1.25	42.1 ^{b} ±3.62	19.1 ^{a} ±0.78	7.6 ^{a} ±0.40	26.7 ^{a} ±1.06
Year							
		2005	20.8 ^{a} ± 1.84	47.7 ^{a} ±4.48	17.4 ^{a} ±2.30	7.5 ^{a} ±0.32	24.9 ^{a} ±2.57
		2006	21.4 ^{a} ± 2.38	42.5 ^{b} ±3.01	17.0 ^{a} ±2.15	7.5 ^{a} ±0.43	24.5 ^{a} ±2.37
Average ± SD			21.1 ± 2.05	45.1 ± 4.55	17.2 ± 2.13	7.5 ± 0.36	24.7 ± 2.37
CV (%)			9.7	10.1	12.4	4.9	9.6

Means (±SD) of triplicate analysis per year and all data on dry weight basis.
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
[1]Biotypes: K = Kabuli, D = Desi. *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles.
[2] Insoluble dietary fiber (IDF), soluble dietary fiber (SDF) and total dietary fiber (TDF)
SD = standard deviation; CV = coefficient of variation

From a nutritional point of view, determination of soluble (SDF), insoluble (IDF) and total dietary fibre (TDF) is important. There was a clear difference in TDF among biotypes, Desi-type chickpea had higher TDF values (26.7%) than did Kabuli-types (22.6%), mainly due to the higher IDF contents of the former (Table 4.1.5). These results coincide with those obtained by Rincon, Martinez, & Ibanez (1998). Gil et al. (1996b) and Rincon et al. (1998) suggested that differences in seed coat thickness between Desi and Kabuli biotypes might have contributed to the differences in TDF and IDF. Seed coat accounts for 14.5-16.5% of seed weight (Paredes-Lopez et al., 1991), and Kabuli-type contains only 4.3% hulls, whereas Desi-type contains 11.5% (Sosulski and Gadan, 1988). On the contrary, very small differences (~0.5%) were found in SDF content (Table 4.1.5). The soluble dietary fibre fraction includes pectins, gums, mucilages and some soluble hemicelluloses (Periago, Ros, Lopez, Martinez & Rincon, 1993). Because hemicellulose constitutes a large proportion (about 55%) of the total dietary fibre of both Desi and Kabuli (Singh, 1984), the SDF content would be independent of seed coat thickness and thus be similar for both biotypes, as revealed in the present results. The 'year' effect was not significant for any of the dietary fibre values.

Correlations between chemical properties are shown in Table 4.1.6. A strong positive correlation was found between IDF and TDF ($r = 0.99$, $P<0.001$). Strong inverse correlations were found between crude fat and protein content ($r = -0.59$, $P<0.01$) as well as total fat and total starch content ($r = -0.72$, $P<0.001$), respectively. Similarly, total starch exhibited very strong negative correlations with TDF and IDF. As anticipated, seed coat amount (SCT) had a positive correlation with dietary fiber, whereas it had a negative relationship with protein, ash and total starch from different chickpea cultivars.

Table 4.1.6 Correlation coefficients (*r*) of chickpea flour chemical composition (combined data, n=12)

	2	3	4	5	6	7	8	9	10
1. Protein	0.56**	-0.59**	-0.02	0.25	0.37	0.47	-0.32	-0.48	-0.60*
2. Ash	1	-0.22	-0.24	0.21	0.38	-0.29	-0.65**	-0.37	-0.56*
3. Crude fat		1	-0.11	-0.28	0.11	0.20	0.32	0.23	0.43
4. Total fat			1	0.09	-0.72***	0.52	0.37	0.53	0.42
5. Amylose				1	-0.23	0.47	0.30	0.46	0.27
6. Total starch					1	-0.77***	-0.34	-0.74***	-0.61*
7. IDF						1	0.60**	0.99***	0.66*
8. SDF							1	0.69*	0.54*
9. TDF								1	0.68*
10.SCT									1

*, **, *** = Significant at p <0.05 0.01 and 0.001, respectively.

IDF: Insoluble dietary fiber, SDF: soluble dietary fiber, TDF: total dietary fiber and SCT: seed coat

4.1.1.4 SDS-PAGE analysis of chickpea flour

SDS-PAGE of chickpea flour under non-reducing conditions is shown in Figure 4.1.3. There was no 'year' effect on the electrophoretograms of chickpea. Kabuli and Desi chickpea contained similar bands in the range of 30 to 55 kDa, with three major bands at approximately 50-55, 40 and 30 kDa. Soy glycinin had major bands at 65, 55, 43, 27, 20 kDa. Soy β-conglycinin had major bands at approximately 50, 30, 27 and 20 kDa. Soy β-conglycinin and glycinin had 55, 27 and 20 kDa bands in common, which might be due to cross contamination during the purification process. Soy and chickpea protein consist of 7S, β-conglycinin, and 11S, glycinin. Altogether these two globulin proteins make up 87 and 57% of the total protein of soybean (Kinsella, 1979) and chickpea (Chavan et al., 1989) protein, respectively. The 11S globulin, which is essentially a hetero-hexamer, contains both acidic and basic subunits with molecular weights in the range of 27 to 37 kDa and 20 to 24 kDa, respectively (Derbyshire, Wright, & Boulter, 1976). On the other hand, 7S globulin, essentially a hetero-trimer, is composed of three discrete protein subunits, α′, α and β-subunits, with molecular weights of 80, 76 and 50 kDa, respectively (Qi, Hettiarchchy, & Kalapathy, 1997). The presence of bands in the range of 30 to 55 kDa confirms the presence of 11S and 7S globulins in the prepared flour samples.

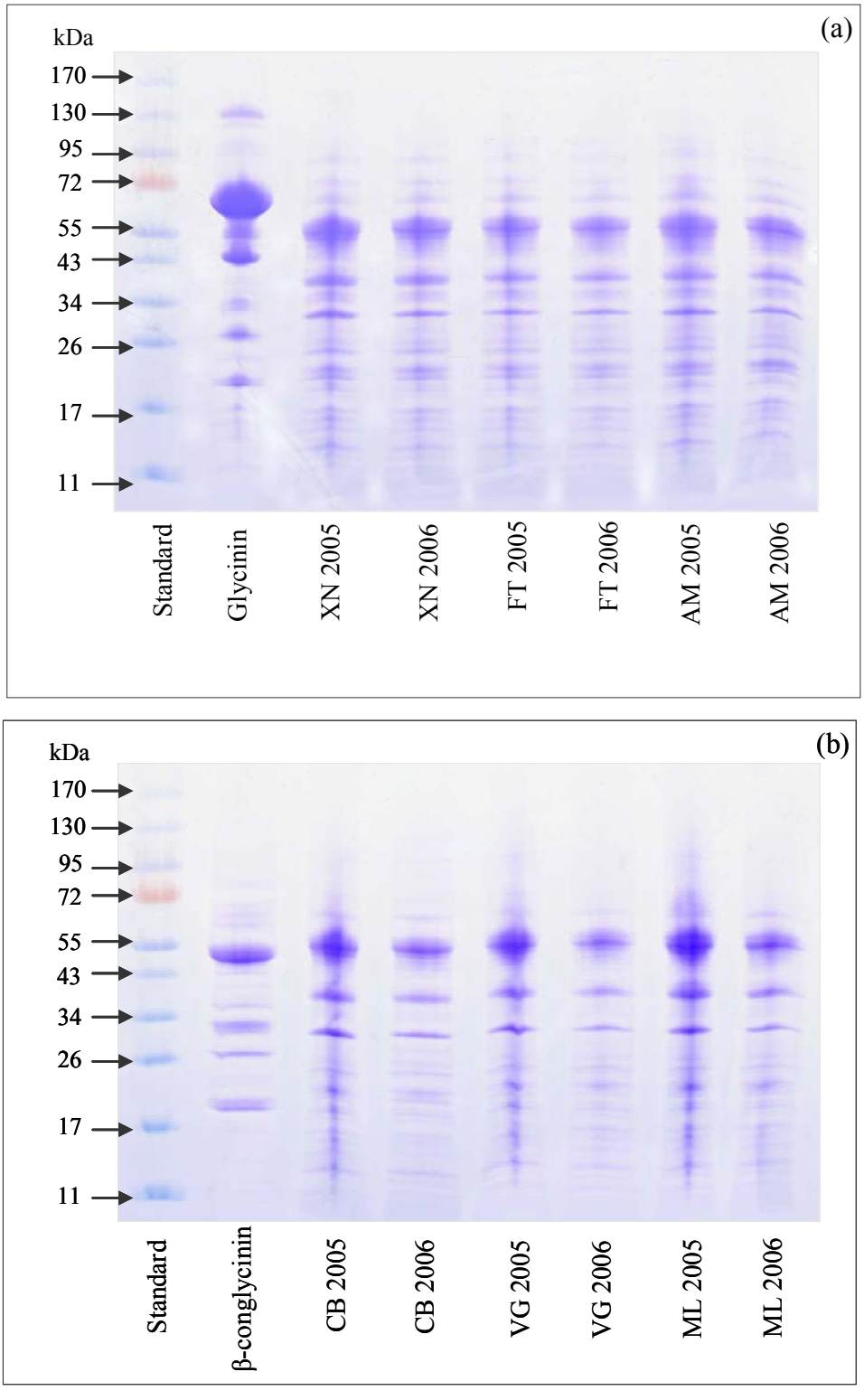

Figure 4.1.3 Electrophoretograms of chickpea flours (a = Kabuli var. and b = Desi var.) and soybean β-conglycinin or gylcinin. Standard (kDa); *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles from 2005 and 2006 harvesting years. 1 µL of the solution (2 µg protein/µL) was loaded.

4.1.1.5 Thermal properties of chickpea flour

4.1.1.5.1 Least gelation concentration

Least gelation concentration (LGC) was taken as a measure of the gelation capacity with the lower the LGC the better the gelation characteristics of the chickpea flour. LGC for various chickpea flours ranged from 6-14% (w/v) (Table 4.1.7). Kabuli chickpea had generally lower LGC (6-10% w/v) than did Desi-types (10-14% w/v). This variation in gelation properties of the two biotypes may be attributed to variation in constituents such as seed coat (Sathe et al., 1982), protein, carbohydrate, and lipids in their flours. CDC Xena and Myles formed very solid and solid gels, respectively, in 20% flour samples. Kaur & Singh (2005) reported 10-14% (w/v) LGC concentrations for various Indian-grown chickpeas. A similar biotype effect on gelation of chickpea flour was also observed (Kaur & Singh, 2005). The LGC reported for other flours, such as cowpea flour, is 16% (w/v) (Abbey & Ibeh, 1991), wing bean flour is 18% (w/v) (Sathe, Deshpande, & Salunkhe, 1982), great northern bean flour is 10% (Sathe & Salunkhe, 1981) and Jackfruit flour is 16% (w/v) (Odoemelam, 2005).

4.1.1.5.2 Thermal stability of chickpea flour

The thermal stability parameters (T_o: onset, T_p: peak temperature and ΔH: enthalpy change) and corresponding DSC thermograms of various chickpea flours are shown in Table 4.1.8 and Figure 4.1.4, respectively. There were two major peaks for chickpea flour, one is a starch gelatinization peak at around 64°C, and there was a second broad peak at a higher temperature (~96°C), which could be interpreted as a protein denaturation peak (Meares, Bogracheva, Hill, & Hedley, 2004). There wasn't a biotype effect for DSC parameters. For starch gelatinization, T_o, T_p and ΔH ranged between 58.13–60.15°C, 63.89–65.35°C and 0.78–1.49 J g^{-1}, respectively. For the protein denaturation peak, the T_o was 88.59–92.63°C, the T_p was 95.11-97.40°C and the ΔH was 0.03–0.11 J g^{-1}.

Table 4.1.7 Least gelation concentration of chickpea flours after heating in boiling water for 1 h followed by cooling for 2 h at 4ºC. (A) Kabuli-types (B) Desi-types.

(A)

Sample concentration (% w/v)	XN		FT		AM	
	gelation	appearance	gelation	appearance	gelation	appearance
2	−	viscous	−	liquid	−	liquid
4	−	viscous	−	liquid	−	liquid
6	+	gel	−	viscous	−	liquid
8	+	gel	−	v. viscous	−	viscous
10	+	firm gel	+	gel	−	viscous
12	+	firm gel	+	gel	+	gel
14	+	v. firm gel	+	firm gel	+	firm gel
16	+	solid gel	+	firm gel	+	firm gel
18	+	v. solid gel	+	v. firm gel	+	firm gel
20	+	v. solid gel	+	v. firm gel	+	v. firm gel

(B)

Sample concentration (% w/v)	CB		VG		ML	
	gelation	appearance	gelation	appearance	gelation	appearance
2	−	liquid	−	liquid	−	viscous
4	−	liquid	−	liquid	−	viscous
6	−	liquid	−	liquid	−	v. viscous
8	−	viscous	−	viscous	+	gel
10	+	gel	+	gel	+	gel
12	+	gel	+	gel	+	firm gel
14	+	firm gel	+	gel	+	firm gel
16	+	firm gel	+	firm gel	+	v. firm gel
18	+	v. firm gel	+	firm gel	+	v. firm gel
20	+	v. firm gel	+	v. firm gel	+	solid gel

Var: XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

Interestingly, there was a 'year' effect with respect to the onset and peak temperatures of the first peak and the onset temperature of second peak (Table 4.1.8). The mean values of the onset and peak temperatures of the first peak had higher values (4.8 and 4.0°C higher, respectively) for the 2006 harvest year than for the 2005 year. On the contrary, as a trend, the second peak (protein denaturation) only had higher onset temperatures (P = 0.11) for chickpea flour obtained from the 2005 harvest compared to flour from the 2006 harvest (2.2°C). Peak temperatures of the second peak were not statistically different.

Table 4.1.8 Year × cultivar, cultivar and year effects on thermal properties of flours from different chickpea cultivars

Var.	year	type	First peak			Second peak		
			T_o (°C)	T_p (°C)	ΔH(J/g)	T_o (°C)	T_p (°C)	ΔH(J/g)
Year × cultivar								
XN	2005	K	57.96 ^{c} ± 0.33	63.34 ^{d} ± 0.04	1.34 ^{abc} ± 0.04	92.77 ^{a} ± 1.42	97.20 ^{ab} ± 1.52	0.06 ^{bcd} ± 0.02
FT	2005	K	55.87 ^{de} ± 0.31	61.67 ^{g} ± 0.32	1.19 ^{bcd} ± 0.03	90.86 ^{ab} ± 4.36	96.46 ^{ab} ± 1.45	0.04 ^{cd} ± 0.02
AM	2005	K	56.58 ^{cde} ± 0.09	62.08 ^{fg} ± 0.19	1.20 ^{bcd} ± 0.07	92.14 ^{a} ± 0.92	96.81 ^{b} ± 0.39	0.06 ^{cd} ± 0.00
CB	2005	D	54.88 ^{e} ± 2.00	62.59 ^{ef} ± 0.57	1.54 ^{abc} ± 0.58	92.28 ^{a} ± 1.47	98.32 ^{a} ± 0.47	0.03 ^{d} ± 0.00
VG	2005	D	56.79 ^{cd} ± 0.22	63.02 ^{de} ± 0.18	0.92 ^{de} ± 0.12	91.14 ^{a} ± 0.33	96.48 ^{b} ± 0.08	0.03 ^{d} ± 0.00
ML	2005	D	55.68 ^{de} ± 1.49	62.84 ^{de} ± 0.81	1.06 ^{cde} ± 0.18	90.24 ^{abc} ± 0.66	97.34 ^{ab} ± 0.98	0.11 ^{a} ± 0.05
XN	2006	K	62.34 ^{a} ± 0.11	67.36 ^{a} ± 0.18	1.64 ^{a} ± 0.14	92.49 ^{a} ± 0.26	94.86 ^{dc} ± 0.42	0.08 ^{abc} ± 0.01
FT	2006	K	60.40 ^{b} ± 0.02	66.11 ^{c} ± 0.00	1.60 ^{ab} ± 0.05	89.54 ^{abc} ± 0.00	98.34 ^{a} ± 0.01	0.04 ^{d} ± 0.00
AM	2006	K	59.94 ^{b} ± 1.41	66.15 ^{bc} ± 0.20	1.73 ^{a} ± 0.19	90.68 ^{abc} ± 0.00	96.25 ^{bc} ± 0.18	0.04 ^{cd} ± 0.00
CB	2006	D	61.39 ^{ab} ± 0.21	66.83 ^{abc} ± 0.25	1.37 ^{ab} ± 0.07	86.72 ^{c} ± 2.87	93.56 ^{d} ± 0.66	0.04 ^{d} ± 0.00
VG	2006	D	61.49 ^{ab} ± 0.06	66.10 ^{c} ± 0.27	0.65 ^{e} ± 0.03	89.79 ^{abc} ± 0.00	93.75 ^{d} ± 0.00	0.05 ^{cd} ± 0.00
ML	2006	D	61.01 ^{ab} ± 0.34	66.90 ^{ab} ± 0.23	1.31 ^{abcd} ± 0.07	86.94 ^{bc} ± 2.85	93.66 ^{d} ± 0.11	0.10 ^{ab} ± 0.03
Cultivar								
XN	05/06	K	60.15 ^{a} ± 3.09	65.35 ^{a} ± 2.84	1.49 ^{a} ± 0.21	92.63 ^{a} ± 0.20	96.03 ^{a} ± 1.65	0.07 ^{b} ± 0.01
FT	05/06	K	58.14 ^{b} ± 3.20	63.89 ^{c} ± 3.14	1.39 ^{a} ± 0.29	90.20 ^{ab} ± 0.93	97.40 ^{a} ± 1.33	0.04 ^{c} ± 0.00
AM	05/06	K	58.26 ^{ab} ± 2.38	64.11 ^{bc} ± 2.88	1.46 ^{a} ± 0.37	91.41 ^{ab} ± 1.03	96.53 ^{a} ± 0.40	0.05 ^{bc} ± 0.01
CB	05/06	D	58.13 ^{b} ± 4.60	64.71 ^{abc} ± 3.00	1.46 ^{a} ± 0.12	89.50 ^{ab} ± 3.93	95.94 ^{a} ± 3.37	0.03 ^{c} ± 0.01
VG	05/06	D	59.14 ^{ab} ± 3.32	64.56 ^{abc} ± 2.18	0.78 ^{b} ± 0.19	90.47 ^{ab} ± 0.95	95.11 ^{a} ± 1.93	0.04 ^{c} ± 0.01
ML	05/06	D	58.34 ^{ab} ± 3.77	64.87 ^{ab} ± 2.87	1.18 ^{ab} ± 0.18	88.59 ^{b} ± 2.34	95.50 ^{a} ± 2.60	0.11 ^{a} ± 0.01
Biotype								
Kabuli			58.85 ^{a} ± 1.07	64.45 ^{a} ± 0.62	1.45 ^{a} ± 0.42	91.41 ^{a} ± 1.88	96.65 ^{a} ± 1.95	0.05 ^{a} ± 0.03
Desi			58.54 ^{a} ± 0.85	64.71 ^{a} ± 0.53	1.14 ^{a} ± 0.39	89.52 ^{a} ± 2.22	95.52 ^{a} ± 1.91	0.06 ^{a} ± 0.03
Year								
2005			56.29 ^{b} ± 2.47	62.59 ^{b} ± 2.40	1.21 ^{a} ± 0.24	91.57 ^{a} ± 1.25	97.10 ^{a} ± 1.15	0.06 ^{a} ± 0.02
2006			61.09 ^{a} ± 3.09	66.57 ^{a} ± 2.10	1.38 ^{a} ± 0.33	89.36 ^{b} ± 2.25	95.07 ^{a} ± 2.12	0.06 ^{a} ± 0.04

Means (±SD) of triplicate analysis per year

Means followed by same letter within column and section do not differ significantly ($P < 0.05$)

[1] Biotypes: K = Kabuli, D = Desi. *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

T_o = onset temperature, T_p = peak temperature and ΔH = enthalpy change of gelatinization or denaturation

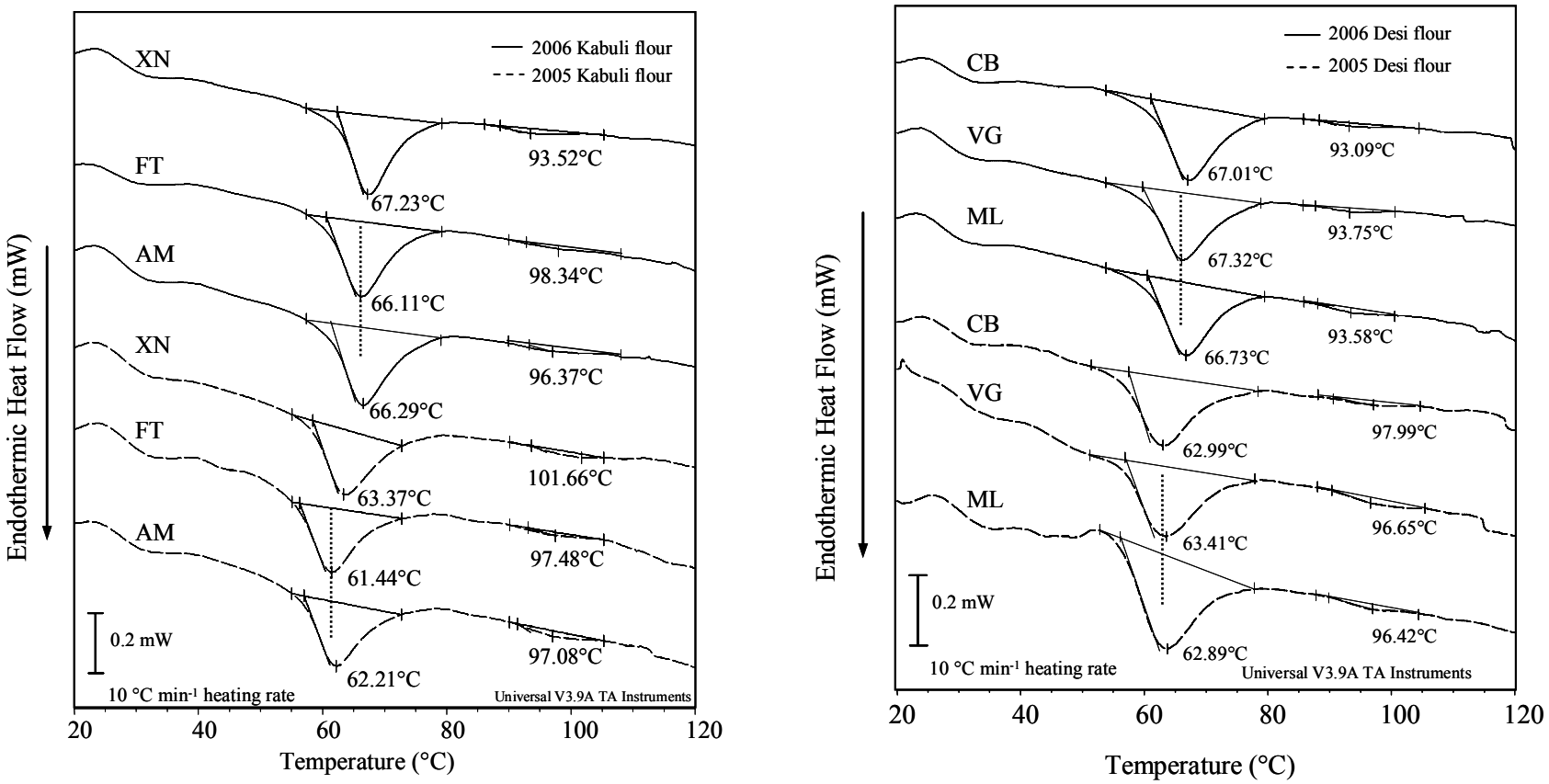

Figure 4.1.4 DSC thermal curves for different chickpea flours from 2005 and 2006 harvesting years. Peak temperatures of the two important peaks are shown. *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

Meares, Bogracheva, Hill, & Hedley (2004) reported average onset and peak temperatures of starch gelatinization for Australian chickpea flour samples of 64 and 72°C, respectively, which are higher than the present results. The thermal properties of chickpea flours from Indian Kabuli and Desi varieties were studied by Kaur & Singh (2005) who also reported higher onset (65.4-67.9°C) and peak (70.6-73.3°C) temperatures than those of the Western Canadian chickpea varieties in our study.

4.1.1.5.3 Pasting properties of chickpea flours

Pasting curves (Figure 4.1.5) and corresponding rapid viscoanalyzer (RVA) parameters (Table 4.1.9) showed significant differences among chickpea cultivars. Pasting temperature (temperature at the onset of the rise in viscosity) of flours from different chickpea cultivars ranged from 61.7 to 68.0°C. Pasting temperature provides an indication of the minimum temperature needed to cook the flour as well as the starch. All flours showed a gradual increase in viscosity with an increase in temperature. The increase in viscosity with temperature is known to be due to the liberation of amylose from the starch granules when they swell (Tester et al., 2004). Final viscosity (indicates the ability of the material to form a viscous paste) and set back (measure of retrogradation tendency or syneresis of flour upon cooling of cooked flour pastes) of chickpea flours ranged from 142.2 to 303.9 and 55.9 to 166.7 RVU, respectively. CDC Frontier had the highest peak (192.2), trough (136.8), breakdown (57.9), final viscosity (303.9) and setback (166.4) viscosities.

For all RVA characteristics a biotype effect was demonstrated. Kabuli-type chickpeas were characterized as having greater average RVA parameters did Desi-types, except for pasting temperature. According to the classification of Schoch & Maywald (1968) based on the pasting profile, chickpea and many other legume starches showed Type C (restricted swelling) viscosity patterns without a pasting peak but with a continual rise throughout the heating period. However, the present study showed Desi-type exhibited viscosity patterns typical of Type

C starches whereas Kabuli-types showed less restricted swelling curves, presenting small pasting peaks, and were more close to the Type B crystalline structure (Figure 4.1.5).

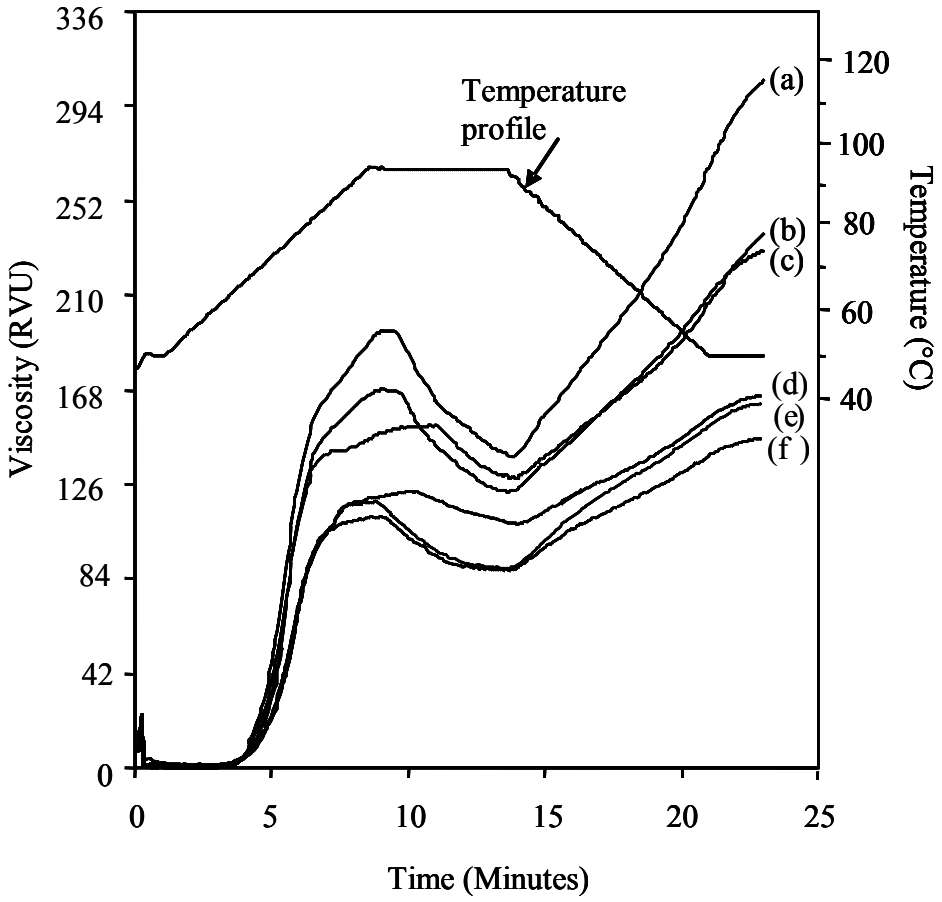

Figure 4.1.5 Rapid visco analyzer pasting profiles of flours (2005) from different chickpea varieties at 14% (w/v) flour: (a) CDC Frontier; (b) CDC Xena, (c) CDC Amit. (d) CDC Cabri; (e) CDC Vanguard and (f) CDC Myles.

Table 4.1.9 Year × cultivar, cultivar and year effects on pasting properties of flours from different chickpea cultivars

Var.	Year	Geno -type[1]	Peak1 (RVU)	Trough1 (RVU)	Breakdown (RVU)	Final viscosity (RVU)	Setback (RVU)	Pasting tem-perature (°C)
Year × cultivar								
XN	2005	K	150.2 ^{d} ± 0.01	127.2 ^{b} ± 0.95	22.3 ^{e} ± 0.01	235.1 ^{b} ± 0.43	106.5 ^{de} ± 1.48	63.6 ^{cd} ± 0.22
FT	2005	K	191.7 ^{a} ± 1.73	136.0 ^{a} ± 1.39	57.1 ^{a} ± 1.63	306.7 ^{a} ± 5.15	163.1 ^{b} ± 4.29	61.7 ^{ef} ± 1.06
AM	2005	K	168.9 ^{b} ± 2.75	123.6 ^{b} ± 2.69	46.7 ^{c} ± 2.11	226.6 ^{b} ± 1.87	105.2 ^{e} ± 1.44	63.0 ^{ed} ± 0.57
CB	2005	D	120.0 ^{e} ± 0.26	103.2 ^{e} ± 4.25	12.6 ^{f} ± 1.48	161.1 ^{d} ± 0.70	54.9 ^{h} ± 0.73	64.5 ^{bc} ± 0.07
VG	2005	D	114.9 ^{f} ± 0.72	87.4 ^{f} ± 1.40	29.0 ^{d} ± 0.01	157.1 ^{d} ± 1.83	71.1 ^{g} ± 1.35	72.0 ^{a} ± 0.02
ML	2005	D	108.3 ^{gh} ± 0.70	84.5 ^{f} ± 0.07	21.7 ^{e} ± 2.15	143.8 ^{ef} ± 0.71	57.9 ^{h} ± 0.18	61.2 ^{f} ± 0.14
XN	2006	K	158.8 ^{c} ± 4.21	126.9 ^{bc} ± 2.30	29.0 ^{d} ± 1.35	228.9 ^{b} ± 4.14	114.3 ^{c} ± 6.00	63.9 ^{cbd} ± 0.11
FT	2006	K	193.0 ^{a} ± 2.69	137.7 ^{a} ± 1.22	58.8 ^{a} ± 0.08	303.0 ^{a} ± 0.23	169.7 ^{a} ± 0.74	61.9 ^{ef} ± 0.04
AM	2005	K	167.0 ^{b} ± 2.12	121.7 ^{c} ± 2.10	52.5 ^{b} ± 3.29	231.3 ^{b} ± 0.71	112.0 ^{cd} ± 5.38	61.8 ^{ef} ± 0.35
CB	2006	D	121.6 ^{e} ± 1.41	113.5 ^{d} ± 4.72	14.3 ^{f} ± 2.16	153.3 ^{ed} ± 3.55	57.1 ^{h} ± 1.63	65.2 ^{b} ± 0.98
VG	2006	D	111.6 ^{gf} ± 0.63	88.1 ^{f} ± 0.67	32.0 ^{d} ± 0.71	176.6 ^{c} ± 16.12	78.8 ^{f} ± 0.30	63.9 ^{cbd} ± 0.15
ML	2006	D	104.9 ^{h} ± 1.24	86.5 ^{f} ± 2.79	24.5 ^{e} ± 0.06	140.5 ^{f} ± 2.88	55.4 ^{h} ± 0.66	62.2 ^{ef} ± 0.40
Cultivar								
XN	05/06	K	154.5 ^{c} ± 5.55	127.0 ^{b} ± 1.44	25.7 ^{d} ± 3.90	232.0 ^{b} ± 4.31	110.4 ^{b} ± 5.75	63.7 ^{bc} ± 0.26
FT	05/06	K	192.2 ^{a} ± 1.93	136.8 ^{a} ± 1.43	57.9 ^{a} ± 1.35	303.9 ^{a} ± 4.43	166.4 ^{a} ± 4.58	61.8 ^{c} ± 0.63
AM	05/06	K	167.9 ^{b} ± 2.28	122.7 ^{b} ± 2.25	49.6 ^{b} ± 4.03	229.0 ^{b} ± 2.95	108.6 ^{b} ± 5.06	62.4 ^{bc} ±0.82
CB	05/06	D	120.8 ^{d} ± 1.24	108.4 ^{c} ± 7.02	13.5 ^{e} ± 1.80	157.2 ^{c} ± 4.95	56.0 ^{d} ± 1.61	64.9 ^{b} ± 0.67
VG	05/06	D	113.2 ^{e} ± 2.01	87.8 ^{d} ± 0.98	30.5 ^{c} ± 1.78	166.9 ^{c} ± 14.63	74.9 ^{c} ± 4.56	68.0 ^{a} ± 4.67
ML	05/06	D	106.6 ^{f} ± 2.13	86.0 ^{d} ± 1.70	23.1 ^{d} ± 2.04	142.2 ^{d} ± 2.56	56.6 ^{d} ± 1.52	61.7 ^{c} ± 0.62
Biotype								
		Kabuli	171.5 ^{a} ± 16.62	128.8 ^{a} ± 6.39	44.4 ^{a} ± 14.60	255.0 ^{a} ± 36.34	128.5 ^{a} ± 28.38	62.6 ^{b} ± 1.02
		Desi	113.6 ^{b} ± 6.27	94.0 ^{b} ± 11.26	22.4 ^{b} ± 7.48	155.4 ^{b} ± 13.39	62.5 ^{b} ± 9.55	64.9 ^{a} ± 3.64
Year								
		2005	142.3 ^{a} ± 31.95	110.5 ^{a} ± 20.56	31.6 ^{b} ± 16.15	205.1 ^{a} ± 59.85	93.1 ^{b} ± 39.12	64.3 ^{a} ± 3.78
		2006	142.8 ^{a} ± 33.60	112.4 ^{a} ± 20.10	35.2 ^{a} ± 16.30	205.3 ^{a} ± 57.64	97.9 ^{a} ± 41.56	63.2 ^{a} ± 1.37

Means (± SD) of triplicate analysis per year.
Means followed by same letter within a column and section do not differ significantly ($P < 0.05$).
[1] Biotypes: K = Kabuli, D = Desi. *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

4.1.1.6 Functional properties of chickpea flours

The functional properties of flours primarily determine their utility in food products. The biotypes belonging to two distinct chickpea groups showed large differences in certain functional properties.

4.1.1.6.1 Nitrogen solubility of chickpea flour

Solubility is probably the most critical of the functional properties of protein, because it affects other properties such as emulsification, foaming and gelation (Kinsella, 1976). The nitrogen solubility profiles of chickpea flours as a function of pH (Figure 4.1.6) indicate minimum solubility (20.3 -35.9%) occurred at pH 4.0-4.4, with an average value of 4.3. These values are similar to those reported for field pea and fababean flour, which were reported to have a minimum nitrogen solubilities of 19.7 and 14.1%, respectively, at pH 4-5 (Sosulski & McCurdy, 1987). Common bean and green mung bean flours had minimum nitrogen solubilities ranging from 18-20% at pH 4.0 (Dzudie & Hardy, 1996).

These soluble proteins are likely albumins. The albumin fraction is less abundant than globulin, and represents 15-25% of the total cotyledonary proteins (Clemente, Vioque, Sánchez-Vioque, Pedroche, Bautista, & Millán, 2000). Except for CDC Xena, all chickpea flours, exhibited high nitrogen solubility values, over 90%, on both sides of the isoelectric pH or 4-5 (Figure 4.1.6). These results closely resemble those reported for field pea and fababean flour (Sosulski & McCurdy, 1987) and common bean and green mung beans flour (Dzudie & Hardy, 1996). In contrast, CDC Xena had low protein solubility (~ 20-30%) over a wider pH range (3.0-4.8). Of particular interest may be the influence of this wide range in protein solubility on the functional properties of meat products when combined.

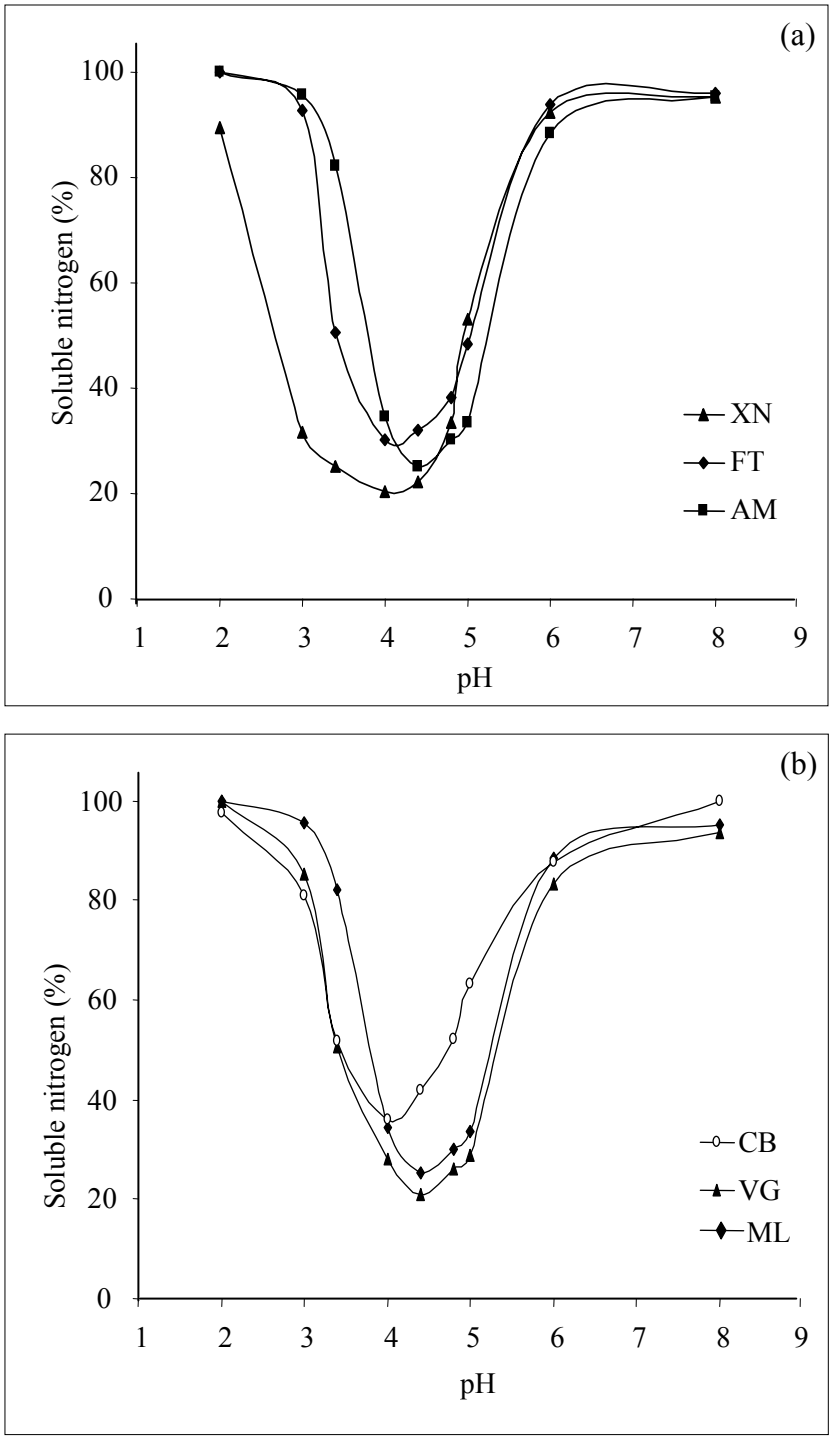

Figure 4.1.6 Nitrogen solubility curves for chickpea flours (2005 harvest) from (a) Kabuli (b) desi var.: XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles as a function of pH.

4.1.1.6.2 Water holding capacity (WHC) and Oil absorption capacity (OAC) of chickpea flours

Kabuli chickpea showed significantly ($P<0.05$) higher WHC/OAC than did Desi (Table 4.1.10). Kaur & Singh (2005) observed a similar biotype effect for OAC, however, they reported an inverse relation for WHC of Kabuli and Desi chickpeas. This discrepancy may be due to varietal differences and methodology differences between the two studies. The water holding and oil absorption capacities of different chickpea varieties from the 2005 and 2006 harvests showed significant differences ($P<0.05$) among cultivars as well as across years (Figure 4.1.6). This year effect may be due to the differences in protein content across the two years (Refer to the Table 4.1.4). However, differences in WHC and OAC for the same cultivar between the two years were not great. WHC and OAC of different cultivars ranged from 0.71 - 0.84 g/g and 0.81 - 0.88 g/g, respectively. CDC Xena had the highest WHC, whereas Myles had the lowest value.

Table 4.1.10 Water holding and oil absorption capacities of flours from different chickpea cultivars

Var.	Year	Biotype[1]	WHC (g/g)	OAC (g/g)
Cultivar				
XN	05/06	K	$0.84^{a} \pm 0.01$	$0.88^{a} \pm 0.08$
FT	05/06	K	$0.79^{b} \pm 0.02$	$0.86^{ab} \pm 0.06$
AM	05/06	K	$0.80^{ab} \pm 0.08$	$0.87^{a} \pm 0.05$
CB	05/06	D	$0.78^{b} \pm 0.00$	$0.78^{c} \pm 0.01$
VG	05/06	D	$0.79^{b} \pm 0.04$	$0.81^{c} \pm 0.06$
ML	05/06	D	$0.71^{c} \pm 0.07$	$0.81^{bc} \pm 0.04$
Biotype				
		Kabuli	$0.81^{a} \pm 0.04$	$0.87^{a} \pm 0.05$
		Desi	$0.76^{b} \pm 0.05$	$0.80^{b} \pm 0.03$
Year				
		2005	$0.81^{a} \pm 0.04$	$0.87^{a} \pm 0.05$
		2006	$0.76^{b} \pm 0.06$	$0.80^{b} \pm 0.04$

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within a column do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi; *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

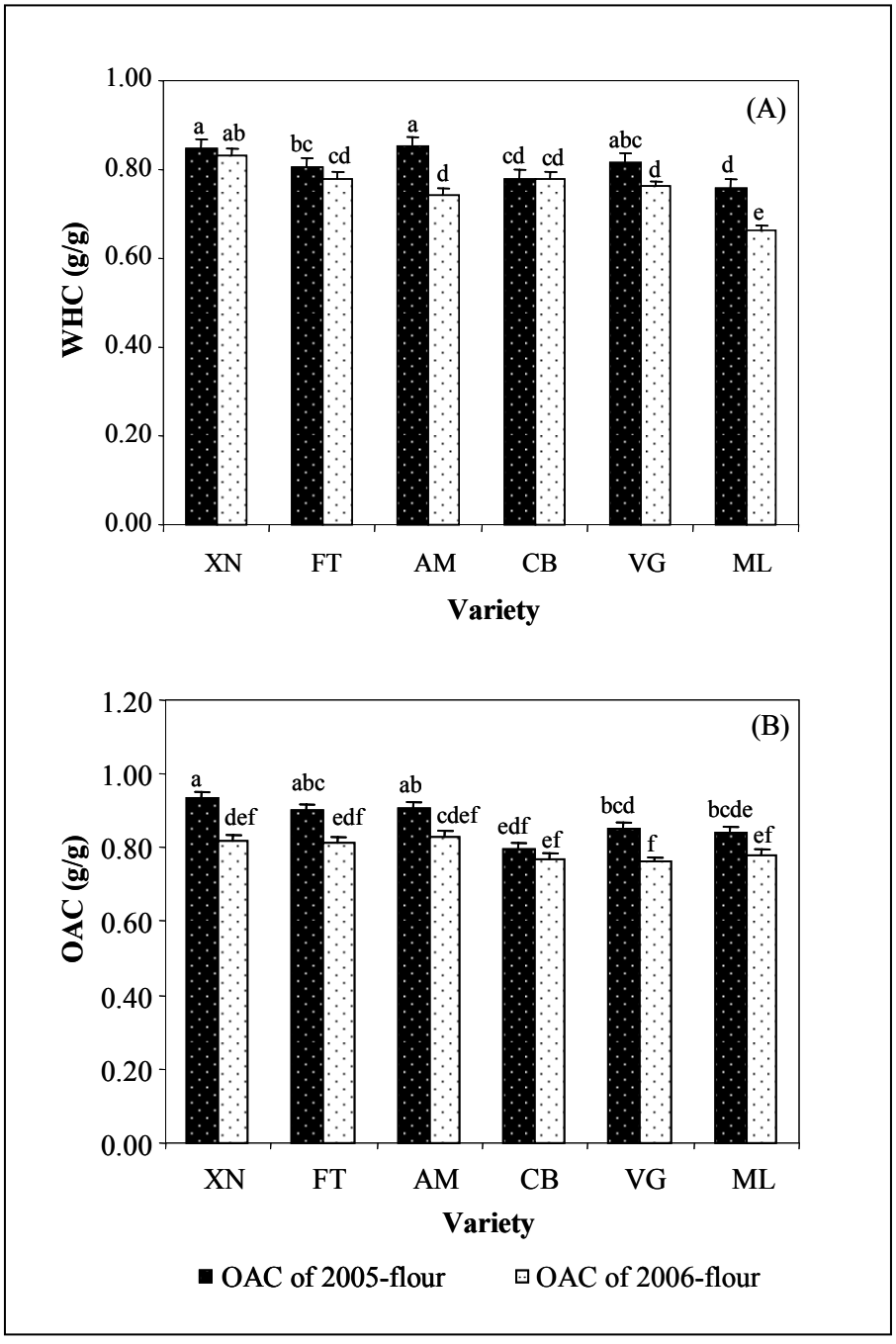

Figure 4.1.7 (A) WHC (B) OAC of chickpea flours from 2005 and 2006 harvesting years. *Var*: XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles. The same letter within WHC or OAC do not differ significantly ($P<0.05$).

Correlations between physical properties of chickpea seed and water and oil holding properties of chickpea flour were investigated (Table 4.1.11). It was evident that several seed properties such as seed volume, hydration capacity and swelling capacity had high positive correlations with WHC and OAC. Seed weight also had a positive correlation with WHC ($P < 0.05$) and OAC ($P < 0.07$). This indicates, bulk properties of chickpea seeds such as SW, SV , HC and SC can be used as predictors of WHC and OAC of flours, which could be useful for plant breeders.

Table 4.1.11 Correlation coefficients (r) of water and oil holding properties of chickpea flours with seed physical properties (combined data, n=12)

	SW	SV	SD	HC	SC
WHC	0.65*	0.76**	-0.32	0.78**	0.77**
OAC	0.54	0.62*	-0.22	0.62*	0.60*

*, ** = Significant at p <0.05 and 0.01, respectively.
Where, WHC-water holding capacity; OAC-oil absorption capacity; SW-seed weight; SV-seed volume; SD-seed density; HC-hydration capacity; SC-swelling capacity

4.1.1.6.3 Emulsion ability index (EAI) and emulsion stability index (ESI)

Emulsion activity is an indicator of how well the flour emulsifies oil, whereas emulsion stability provides information of strength of the intact emulsion over time. Emulsifying properties of chickpea flours from the two harvests are shown in Figure 4.1.8. Cultivar, biotype and year effects on EAI and ESI of chickpea flours are shown in Table 4.1.12. The Kabuli-type flours had higher emulsifying properties than did Desi-type flour again proving their higher quality. This might be because flours from Kabuli-types had significantly higher ($P < 0.05$) protein contents than did flours from Desi-types. Other than Myles, all varieties showed minor differences in EAI and ESI among chickpea cultivars from the two different years. Among the Kabuli biotype, CDC Xena and CDC Amit had the highest emulsion activities of 0.43 and 0.40, respectively, but did not form as stable emulsion as CDC Frontier. In the case of Desi-type, Myles had superior EAI and ESI compared to other Desi varieties (0.41 and 21.5, respectively).

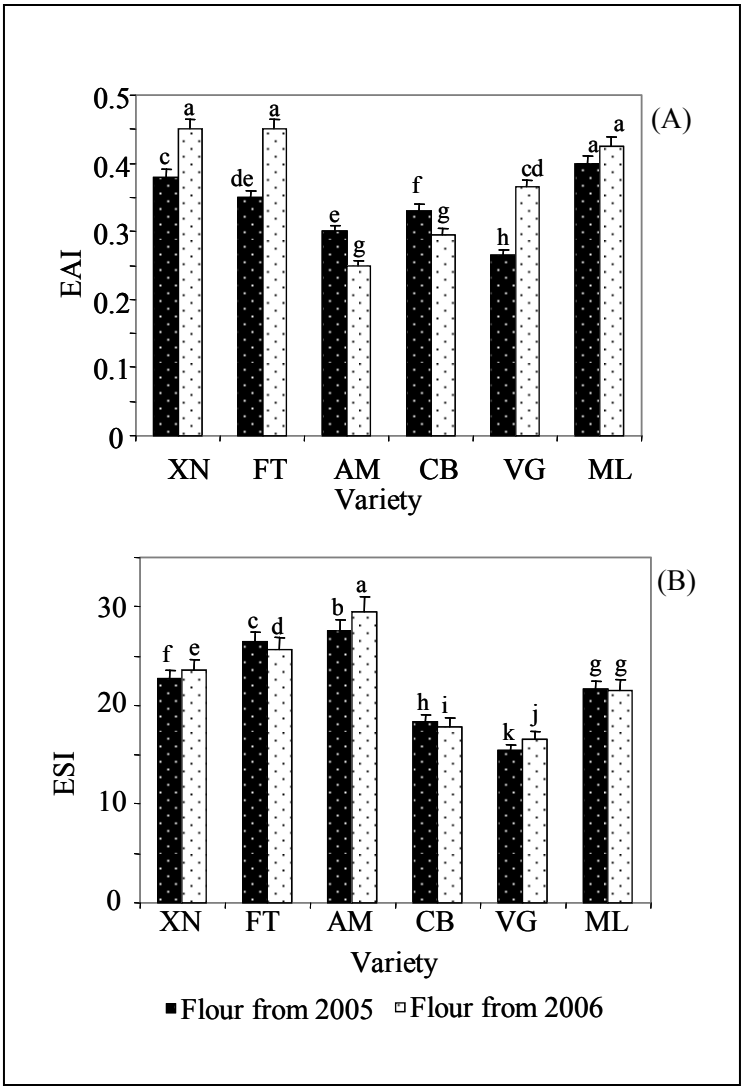

Figure 4.1.8 (A) EAI (B) ESI of chickpea flours from 2005 and 2006 harvesting years. *Var*: XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles. The same letter within WHC or OAC do not differ significantly ($P < 0.05$).

Table 4.1.12 Emulsion properties of different chickpea flours

Var.	Year	Biotype[1]	EAI	ESI
Cultivar				
XN	05/06	K	0.43 [a] ± 0.05	23.00 [c] ± 0.50
FT	05/06	K	0.40 [a] ± 0.07	26.08 [b] ± 0.46
AM	05/06	K	0.28 [b] ± 0.04	28.55 [a] ± 1.34
CB	05/06	D	0.30 [b] ± 0.02	18.05 [d] ± 0.35
VG	05/06	D	0.32 [b] ± 0.07	15.95 [e] ± 0.78
ML	05/06	D	0.41 [a] ± 0.02	21.5 [c] ± 0.07
Biotype				
		Kabuli	0.38 [a] ± 0.18	25.88 [a] ± 13.86
		Desi	0.34 [b] ± 0.17	18.52 [b] ± 10.08
Year				
		2005	0.34 [a] ± 0.02	21.99 [a] ± 0.44
		2006	0.37 [a] ± 0.06	22.40 [a] ± 4.75

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within a column do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi; *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

Except in a few cases, significant correlations among chemical, thermal and functional properties of chickpea flours were not observed (data not shown). However, there was a positive correlation between total starch and onset temperature of DSC data (peak one, corresponding to the gelatinization of starch in chickpea flour) as well as peak value of RVA data (r = 0.63, $P < 0.05$; r = 0.69, $P < 0.05$, respectively). Furthermore, TDF had a strong negative relationship with RVA peak value ($r = -0.91$, $P < 0.001$).

4.1.1.7 Summary and conclusions

Flours from Kabuli and Desi chickpea cultivars differed significantly in most of their physico-chemical, thermal and functional properties. CDC Xena seeds had the highest seed weight, hydration capacity, hydration index, swelling capacity and swelling index. Biotype

statistically affected the seed weight, seed coat, hydration capacity and swelling capacity, with the mean values of Kabuli-types higher than those of Desi-types except for seed coat. The high positive and significant correlations between seed size (weight and volume) and hydration or swelling capacities offers breeders the possibility of indirect selection for these two quality characters by selecting for seed size. Kabuli chickpea seeds and flour were lighter in colour than Desi seeds or flour, but the opposite relationship showed for redness. Chemical composition of flour showed that Kabuli-type flour had higher total starch and protein content but lower amylose, IDF and TDF values than did Desi-types. The amount of seed coat and IDF or TDF had a direct relationship. Kabuli-type chickpea, having high seed weight, hydration capacity, swelling capacity and protein content, had higher water holding and oil absorption capacities than did Desi-types. SDS-PAGE showed major bands of the subunits of the 7S and 11S globulin, suggesting primarily that these proteins are present in the flour.

CDC Xena (the lowest value among the Kabuli-type) produced a gel at a flour concentration as low as 6% (w/v), whereas Myles (the lowest value of the Desi-type) gelled at 8% (w/v) concentration. In DSC thermograms of chickpea flour, two peaks were evident, which corresponded to starch gelatinization and protein denaturation. Even though DSC parameters did not show biotype effects, it reflected the year effect by having higher onset and peak temperatures (approximately 4°C higher) for starch gelatinization for samples from the 2006 harvest. On the contrary, pasting properties of flours from Kabuli- and Desi-types were significantly different. Protein in flour from CDC Xena were relatively insoluble over a wide range of pH (3-5).

Protein plays a key role in WHC, OAC, EAI and ESI of chickpea flour. Having higher protein contents, Kabuli-types had higher values for these functional properties than Desi-types typically had. Now that we understand more about the flour, compositional, thermal and functional properties of chickpea flour, the next step is to understand the nutritional and anti-nutritional properties of flour from Western Canadian chickpea cultivars.

4.1.2 Chemical, functional and thermal properties of protein isolates from six Western Canadian chickpea (*Cicer arietinum* L.) cultivars.

4.1.2.1 Proximate analysis of chickpea protein isolates

The proximate composition of the protein isolates from different cultivars is presented in Table 4.1.13. Except for crude fat content, other chemical composition parameters did not significantly differ among biotypes. However, the Desi-type protein isolates had significantly higher crude fat contents than did the Kabuli-type (>0.8%). Analysis of chemical properties of chickpea protein isolates between years did not show any differences, except that 2006 protein isolates had slightly higher ash contents than those prepared from 2005 cultivars. Protein isolates prepared from different chickpea cultivars exhibited protein contents of 72.8-85.3%. Soy protein isolates (SPI) prepared in a similar manner had higher protein contents (88.9%) than did the chickpea protein isolates (Table 4.1.13), whereas pea protein isolates (PPI) had protein levels similar to the chickpea isolates (Table 4.1.14). Mean ash contents of chickpea isolates ranged from 3.2 to 4.5%. Pea protein isolate had the highest ash content of 5.9%. Ash contents of 2.9% in chickpea protein isolates (Sánchez-Vioque et al., 1999a) and 4.6% in winged bean (Sathe et al., 1982b) and 0.71% in lupin seed protein concentrates (Sathe et al., 1982a) have been reported.

Analysis also showed a low level of starch (0.2 - 0.5%) in the chickpea protein fractions. However, protein isolates from different chickpea cultivars contained a relatively high amount of crude fat (7.4 - 9.3%) and total lipids (11.4 - 14.8%). Fat contents of 9.0% for winged bean (Sathe et al., 1982b) and 17.9% for lupin protein concentrates (Sathe et al., 1982a) have been reported. Soy protein isolate and PPI had relatively low crude fat and total lipid contents (Table 4.1.14). This is because SPI and PPI that we used were defatted samples.

Table 4.1.13 Year × cultivar, cultivar, year and biotype effects on chemical composition of protein isolates on dry weight basis

Var.	Year	Geno-type[1]	Protein[2] (%)	Ash (%)	Crude-fat (%)	Total fat (%)	Total starch (%)
Year × cultivar							
XN	2005	K	81.5 ^{c} ± 0.04	3.5 ^{f} ± 0.05	7.7 ^{i} ± 0.02	13.9 ^{ed} ± 0.07	0.2 ^{ef} ± 0.00
FT	2005	K	75.5 ^{e} ± 0.15	4.2 ^{cd} ± 0.07	8.2 ^{d} ± 0.03	12.8 ^{g} ± 0.10	0.5 ^{c} ± 0.01
AM	2005	K	76.8 ^{d} ± 0.05	3.5 ^{f} ± 0.00	8.0 ^{gf} ± 0.02	13.6 ^{a} ± 0.30	0.2 ^{ef} ± 0.00
CB	2005	D	73.0 ^{g} ± 0.05	4.1 ^{d} ± 0.07	9.5 ^{c} ± 0.01	12.2 ^{h} ± 0.29	0.4 ^{d} ± 0.00
VG	2005	D	76.7 ^{d} ± 0.03	3.1 ^{g} ± 0.00	9.8 ^{a} ± 0.03	15.4 ^{c} ± 0.02	0.2 ^{ef} ± 0.00
ML	2005	D	82.1 ^{b} ± 0.07	3.8 ^{e} ± 0.00	7.6 ^{j} ± 0.03	13.2 ^{gf} ± 0.03	0.7 ^{a} ± 0.02
XN	2006	K	89.1 ^{a} ± 0.25	3.5 ^{f} ± 0.08	7.5 ^{gh} ±0.01	14.2 ^{d} ± 0.07	0.2 ^{e} ± 0.00
FT	2006	K	73.9 ^{f} ± 0.01	4.9 ^{a} ± 0.09	8.6 ^{e} ±0.10	13.6 ^{ef} ± 0.40	0.3 ^{e} ± 0.01
AM	2006	K	75.6 ^{e} ± 0.04	3.9 ^{e} ± 0.06	7.9 ^{f} ±0.10	10.5 ^{i} ± 0.91	0.6 ^{b} ± 0.05
CB	2006	D	72.6 ^{gh} ± 0.40	4.7 ^{b} ± 0.09	9.1 ^{b} ±0.14	10.5 ^{i} ± 0.31	0.4 ^{d} ± 0.00
VG	2006	D	72.4 ^{h} ± 0.36	3.3 ^{g} ± 0.04	9.8 ^{a} ± 0.07	13.7 ^{edf} ± 0.15	0.2 ^{f} ± 0.02
ML	2006	D	77.2 ^{d} ± 0.67	4.3 ^{c} ± 0.16	7.1 ^{hi} ± 0.12	16.4 ^{b} ± 1.19	0.2 ^{e} ± 0.00
Cultivar							
XN	05/06	K	85.3 ^{a} ± 4.42	3.5 ^{cd} ± 0.03	7.6 ^{c} ± 0.15	12.3 ^{a} ± 0.21	0.2 ^{a} ± 0.00
FT	05/06	K	74.7 ^{b} ± 0.95	4.6 ^{a} ± 0.54	8.4 ^{b} ± 0.27	13.2 ^{a} ± 0.57	0.4 ^{a} ± 0.14
AM	05/06	K	76.2 ^{b} ± 0.71	3.7 ^{bc} ± 0.27	7.9 ^{bc} ± 0.05	12.1 ^{a} ± 2.19	0.4 ^{a} ± 0.28
CB	05/06	D	72.8 ^{b} ± 0.34	4.4 ^{a} ± 0.44	9.3 ^{a} ± 0.27	11.4 ^{a} ± 1.20	0.4 ^{a} ± 0.00
VG	05/06	D	74.5 ^{b} ± 2.48	3.2 ^{d} ± 0.11	9.8 ^{a} ± 0.02	14.5 ^{a} ± 1.20	0.2 ^{a} ± 0.00
ML	05/06	D	79.6 ^{ab} ± 2.89	4.1 ^{ab} ± 0.34	7.4 ^{c} ±0.36	14.8 ^{a} ± 2.26	0.5 ^{a} ± 0.35
Biotype							
		Kabuli	78.7 ^{a} ± 5.46	3.9 ^{a} ± 0.56	8.0 ^{b} ± 0.36	13.4 ^{a} ± 3.42	0.3 ^{a} ± 0.16
		Desi	75.7 ^{a} ± 3.63	3.9 ^{a} ± 0.63	8.8 ^{a} ± 1.16	13.3 ^{a} ± 2.14	0.3 ^{a} ± 0.20
Year							
		2005	77.6 ^{a} ± 3.37	3.7 ^{b} ± 0.41	8.5 ^{a} ± 1.01	13.5 ^{a} ± 3.14	0.4 ^{a} ± 0.21
		2006	76.8 ^{a} ± 6.04	4.1 ^{a} ± 0.67	8.3 ^{a} ± 0.92	13.2 ^{a} ± 2.90	0.3 ^{a} ± 0.16
Average [3] ± SD			77.2 ± 4.90	3.9 ± 0.57	8.39 ± 0.93	13.3 ± 1.70	0.32 ± 0.18
CV (%)			6.35	14.58	11.04	9.50	55.74

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi , [2]Total nitrogen × 6.25
Var: XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles
[3] Average of properties of chickpea proteins; SD = standard deviation; CV = coefficient of variation

Table 4.1.14 Chemical composition of soy and pea protein isolates on dry weight basis

Protein	Protein[1] (%)	Ash (%)	Crude fat (%)	Total fat (%)	Total starch (%)
SPI	88.9 ± 1.34	4.2 ± 0.14	0.5 ± 0.14	3.8 ± 0.07	0.7 ± 0.21
PPI	79.6 ± 0.14	5.9 ± 0.07	2.4 ± 0.21	8.0 ± 0.14	1.0 ± 0.01

Means (±SD) of triplicate analysis and all data on dry weight basis
[1]Total nitrogen × 6.25

4.1.2.2 Colour characteristics of protein isolates

Due to complete removal of the seed coat from chickpea seed prior to protein separation (see Figure 3.1 in section 3.0), there was minimal contamination of chickpea protein isolates with seed coat material. This was confirmed by Kabuli and Desi protein isolates having the same L*, a* and b* values (Table 4.1.15). However, protein isolates from the 2006 harvest had higher a* values than did protein isolates from the 2005 harvest, indicating that protein isolates from 2006 seeds were more red in colour than were corresponding 2005 protein isolates. CIE colour values of different chickpea isolates were significantly different for redness and yellowness values. PPI had the highest L*, ΔE and a* values, indicating it was lighter and redder in colour than other samples (Table 4.1.15). SPI was also lighter than the chickpea isolates (L* and ΔE).

In order to study the effect of fat on colour, the flour was defatted by the method of Folch, Lees, & Stanley (1957) as modified for plant materials by Christie (1993) (data not shown). It was observed that removal of fat from protein isolates increased the lightness and decreased redness and yellowness (mean L* value from 73.18 → 84.82; a* value from 3.92 → 0.66; b* value from 25.49 → 12.14) of the samples, which was in accordance with visual observation. This may be due to removal of oil soluble pigments from chickpea flour and protein isolates.

Table 4.1.15 Year × cultivar, cultivar and year effects on CIE colour values of protein isolates from different chickpea cultivars

Var.	year	type[1]	*L**	*a**	*b**	Δ*E*
Year × cultivar						
XN	2005	K	76.22 ^{b} ± 0.17	2.01 ^{k} ± 0.02	21.53 ^{h} ± 0.14	14.89 ^{e} ± 0.09
FT	2005	K	74.31 ^{c} ± 0.19	3.12 ^{g} ± 0.03	23.96 ^{c} ± 0.07	18.14 ^{f} ± 0.21
AM	2005	K	73.18 ^{e} ± 0.17	3.55 ^{f} ± 0.05	26.07 ^{d} ± 0.26	20.95 ^{h} ± 0.28
CB	2005	D	73.81 ^{d} ± 0.40	2.25 ^{j} ± 0.05	21.64 ^{h} ± 0.36	10.75 ^{b} ± 0.42
VG	2005	D	73.57 ^{de} ± 0.16	2.79 ^{I} ± 0.02	22.87 ^{g} ± 0.19	13.83 ^{d} ± 0.50
ML	2005	D	74.44 ^{c} ± 0.23	2.99 ^{h} ± 0.02	24.94 ^{e} ± 0.02	20.10 ^{g} ± 0.48
XN	2006	K	77.43 ^{a} ± 0.05	2.86 ^{I} ± 0.01	18.49 ^{I} ± 0.09	12.92 ^{c} ± 0.12
FT	2006	K	70.74 ^{g} ± 0.59	6.44 ^{a} ± 0.15	30.69 ^{b} ± 0.49	31.11 ^{j} ± 0.14
AM	2006	K	71.13 ^{gf} ± 0.26	6.32 ^{b} ± 0.03	31.58 ^{a} ± 0.09	33.43 ^{k} ± 0.37
CB	2006	D	76.17 ^{b} ± 0.01	3.90 ^{e} ± 0.06	26.45 ^{c} ± 0.17	28.41 ^{I} ± 0.44
VG	2006	D	71.48 ^{f} ± 0.02	5.55 ^{c} ± 0.02	31.89 ^{a} ± 0.02	33.21 ^{k} ± 0.11
ML	2006	D	65.66 ^{h} ± 0.03	5.20 ^{d} ± 0.03	25.75 ^{d} ± 0.13	8.57 ^{a} ± 0.23
Cultivar						
XN	05/06	K	76.83 ^{a} ± 0.67	2.44 ^{c} ± 0.47	20.01 ^{b} ± 1.67	13.91 ^{a} ± 1.08
FT	05/06	K	72.53 ^{a} ± 1.96	4.78 ^{a} ± 1.82	27.33 ^{ab} ± 3.69	24.63 ^{a} ± 7.11
AM	05/06	K	72.16 ^{a} ± 1.13	4.94 ^{a} ± 1.52	28.83 ^{a} ± 3.02	27.19 ^{a} ± 6.84
CB	05/06	D	74.99 ^{a} ± 1.35	3.08 ^{bc} ± 0.91	24.05 ^{ab} ± 2.65	19.58 ^{a} ± 9.67
VG	05/06	D	72.53 ^{a} ± 1.17	4.17 ^{ab} ± 1.51	27.38 ^{ab} ± 4.95	23.52 ^{a} ± 10.62
ML	05/06	D	70.05 ^{a} ± 4.81	4.10 ^{ab} ± 1.21	25.35 ^{ab} ± 0.46	14.34 ^{a} ± 6.32
SPI			83.71 ± 0.11	6.18 ± 0.01	17.73 ± 0.03	30.59 ± 0.16
PPI			86.52 ± 0.42	8.06 ± 0.07	20.05 ± 0.32	44.60 ± 1.61
Biotype						
		Kabuli	73.84 ^{a} ± 2.53	4.05 ^{a} ± 1.76	25.39 ^{a} ± 5.64	21.91 ^{a} ± 5.23
		Desi	72.52 ^{a} ± 3.47	3.78 ^{a} ± 1.27	25.59 ^{a} ± 5.71	19.15 ^{a} ± 5.26
Year						
		2005	74.26 ^{a} ± 17.06	2.79 ^{b} ± 0.82	23.50 ^{a} ± 5.64	16.44 ^{a} ± 5.23
		2006	72.10 ^{a} ± 3.98	5.05 ^{a} ± 3.98	27.48 ^{a} ± 5.71	20.61 ^{a} ± 5.26
Average [2] ± SD			73.18 ± 3.07	3.92 ± 1.64	25.49 ± 4.37	20.53 ± 5.53
CV (%)			4.20	41.86	17.16	26.96

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within column and section do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi, *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles
[2] Average of properties of chickpea proteins; SD = standard deviation; CV = coefficient of variation

4.1.2.3 SDS-PAGE of protein isolates under non-reducing conditions

The chickpea globulins, the main storage proteins, are mainly constituted by 11S legumins and 7S vicilins, which represent 60-80% of the extractable protein (Clemente et al., 2000). The SDS-PAGE patterns for Kabuli- and Desi-type protein isolates from two different harvesting years were basically identical (Figure 4.1.9). Purified soy β-conglycinin (7S) and glycinin (11S) and chickpea isolates from 2005 and 2006 harvests were routinely compared for SDS-PAGE polypeptide profile under non-denaturing conditions. Soy glycinin had at least six polypeptide bands in the range of 20-34 kDa and 43-72 kDa, with two main protein bands at 43 kDa and ~ 66 kDa. Chickpea protein isolates also had a band at ~ 66 kDa. A similar molecular mass band, which was identified as a monomer of 11S globulin, was also reported in previous studies at 66.2 kDa for SPI (Horax, Hettiarchchy, Chen, & Jalaluddin, 2004), at 66.4 kDa for chickpea protein (Paredes-Lopez et al., 1991) and at 64 kDa for red bean legumin (Meng & Ma, 2002) under non-reducing condition. The quaternary structure of legumin in legume seeds is made up of essentially six such subunits. Soy β-conglycinin had a major band at 54 kDa and four minor bands at 20, 28, 32 and 35 kDa. Similarly, chickpea proteins had corresponding bands at 31, 38 and 55 kDa. Bands at ~20, 30-35 and 55 kDa are typical for the subunits of 7S vicilin protein (Koyoro & Powers, 1987; Meng & Ma, 2002; Rangel, Domont, Pedrosa, & Ferreira, 2003). However, these and several other polypeptides are also found in the soy glycinin fraction. This may be due to the cross-contamination of each fraction prepared by isoelectric precipitation.

Banding patterns of chickpea isolates had close resemblance to the molecular band patterns of the chickpea flour (see Figure 4.1.3).

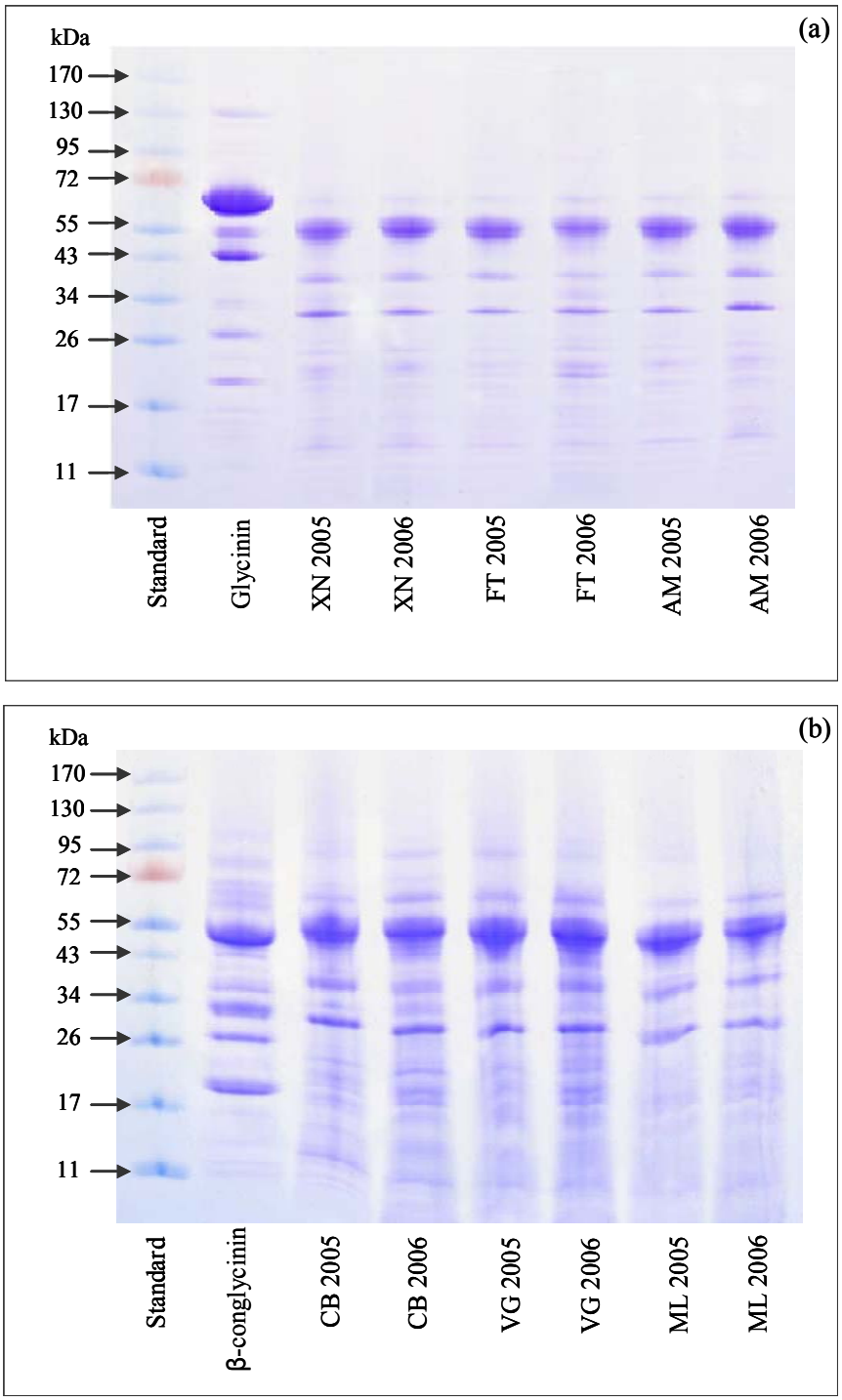

Figure 4.1.9 Electrophoretograms of chickpea (a = Kabuli var. and b = desi var.) protein isolates and soybean conglycinin or gylicinin. Standard (kDa); *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles from 2005 and 2006 harvesting years.

4.1.2.4 Thermal properties of chickpea protein isolates

4.1.2.4.1 Least gelation concentration (LGC)

The ability of plant proteins to form thermally-induced gels is essential with respect to their use in food systems. Gel formation ability of proteins in food systems is very important due to their ability to hold water, lipids, sugars, flavours and other ingredients (Kinsella, 1979). LGC for various protein isolates ranged from 10-14% (Table 4.1.16). Generally, chickpea flour had lower LGCs than did the chickpea protein isolates, possibly due to starch gelatinization in the flour. CDC Xena, Myles and SPI formed a gel when protein concentrates were as low as 10%. PPI had LGC at 14% protein concentration. Kaur & Singh (2006) reported 14-18% LGC for various Indian chickpea isolates. LGC of 12% for cowpea (Horax, Hettiarchchy, Chen, & Jalaluddin, 2004), 14% for winged bean (Sathe, Deshpande, & Salunkhe, 1982b) and, 8% for lupin (Sathe, Deshpande, & Salunkhe, 1982a) and great northern bean protein (Sathe & Salunkhe, 1981) have been reported.

4.1.2.4.2 Gelation of chickpea proteins (rheological properties)

Figure 4.1.10 A & B shows the structural development of chickpea protein slurries during heating and cooling as a function of time and temperature, respectively. Onset temperature (or gelation temperature) for structure development of chickpea protein was defined as the G′-G″ crossover point. Onset temperature of different chickpea proteins varied between 62.4 and 78.7°C (Figure 4.1.10 A: inset). Protein isolates from Kabuli-type chickpea generally had lower onset temperatures (ranged from 62.4 to 67.2°C) than did those for Desi-types (ranged from 66.4 to 78.7°C). Final gel strength was also remarkably different among the different cultivars across years (Table 4.1.17). Myles had the second highest protein conent and had the heighest G′ value (2735.5 Pa) for chickpea isolates made from the 2005/2006 harvests, whereas the lowest G′ (813.2 Pa) was observed for CDC Cabri (with the lowest protein content) for 2005/2006.

Table 4.1.16 Least gelation concentration of chickpea isolates after heating in boiling water for 1h followed by cooling for 2 h at 4 ºC. (A) Kabuli-types (B) Desi-types (C) SPI and PPI.

(A)

Sample con-centration (% w/v)	XN		FT		AM	
	gelation	appearance	gela-tion	appearance	gela-tion	appearance
2	−	liquid	−	liquid	−	liquid
4	−	liquid	−	liquid	−	liquid
6	−	viscous	−	liquid	−	liquid
8	−	v. viscous	−	viscous	−	viscous
10	+	gel	−	viscous	−	viscous
12	+	gel	−	v. viscous	−	viscous
14	+	firm gel	+	gel	+	gel
16	+	firm gel	+	gel	+	gel
18	+	solid gel	+	gel	+	gel
20	+	solid gel	+	gel	+	firm gel

(B)

Sample concen-tration (% w/v)	CB		VG		ML	
	gelation	appearance	gela-tion	appearance	gela-tion	appearance
2	−	liquid	−	liquid	−	viscous
4	−	liquid	−	liquid	−	viscous
6	−	liquid	−	viscous	−	v. viscous
8	−	viscous	−	v. viscous	−	v. viscous
10	−	v. viscous	−	v. viscous	+	gel
12	−	v. viscous	+	gel	+	firm gel
14	+	gel	+	v. firm gel	+	v. firm gel
16	+	firm gel	+	v. firm gel	+	solid gel
18	+	firm gel	+	solid gel	+	v. solid gel
20	+	v. firm gel	+	solid gel	+	v. solid gel

(C)

Sample concentration (% w/v)	SPI		PPI	
	gelation	appearance	gelation	appearance
2	−	liquid	−	liquid
4	−	liquid	−	liquid
6	−	viscous	−	viscous
8	−	v.viscous	−	v. viscous
10	+	gel	−	v. viscous
12	+	gel	−	Weak gel
14	+	firm gel	+	gel
16	+	firm gel	+	gel
18	+	solid gel	+	v. firm gel
20	+	v. solid gel	+	v. firm gel

Var: XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles, SPI = soy protein isolates, PPI = pea protein isolates

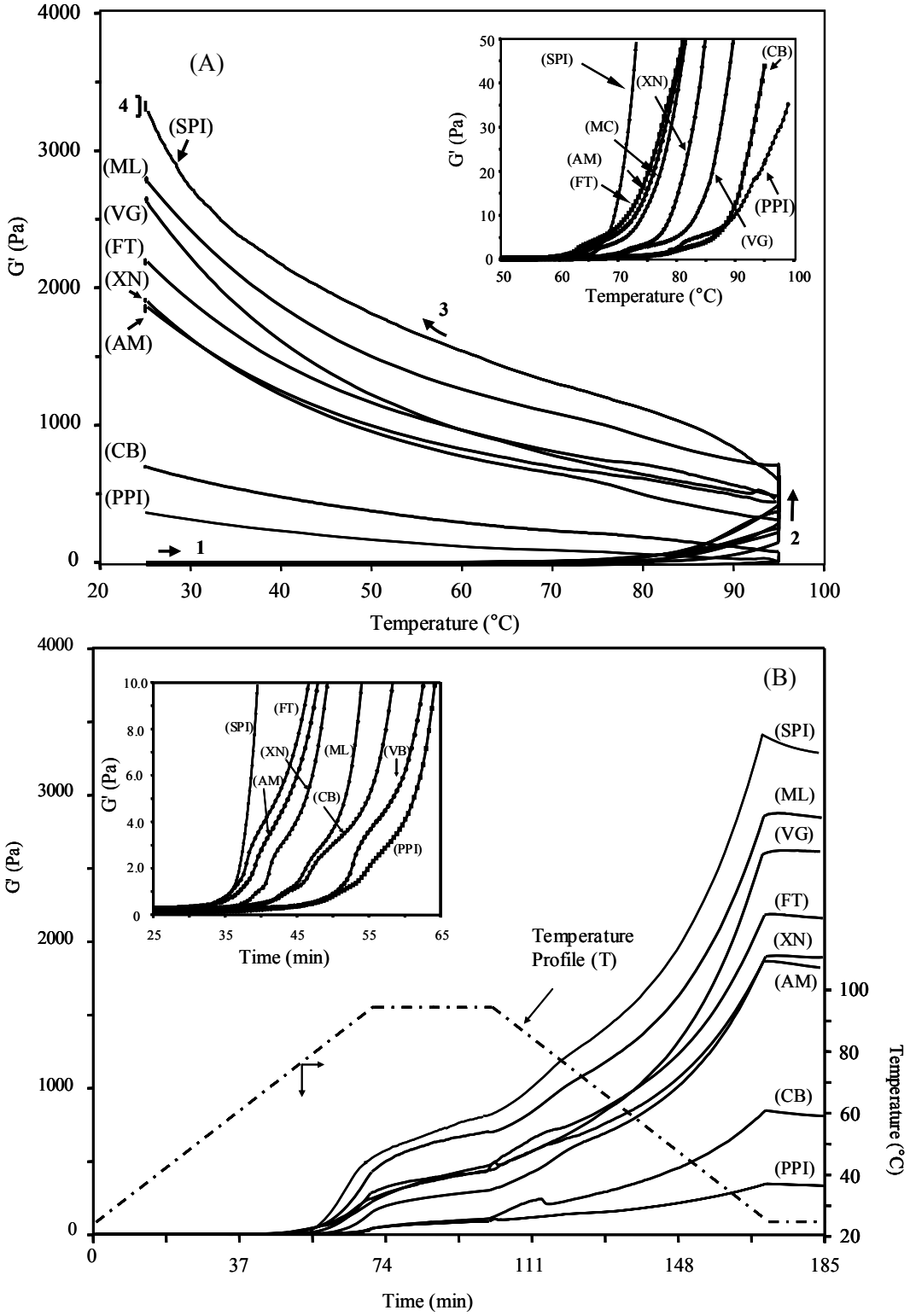

Figure 4.1.10 Storage (*G'*) moduli of 12% chickpea protein isolate dispersion in deionized water at pH 7.0 as a function of (A) temperature and (B) time during heating and cooling cycle (*T*): (FT) CDC Frontier; (AM) Amit; (ML) Myles; (XN) CDC Xena; (VG) CDC Vanguard; (CB) CDC Cabri; (SPI) soy protein isolates (PPI) pea protein isolates *Insets:* Storage (*G'*) modulus during heating phase. Heating and cooling rate was 1 K/min.

Table 4.1.17 Storage modulus of different chickpea isolates (12% w/w on protein basis) at the end of heating and cooling process

Cultivar	Year	Bio-type[1]	G′ (Pa)	G″ (Pa)	Tan δ	Onset tem-perature (°C)
Year × cultivar						
XN	2005	K	1360.0 ^{h} ± 14.1	226.0 ^{h} ± 7.1	0.188 ^{b} ± 0.0	66.9 ^{c} ± 0.2
FT	2005	K	2263.5 ^{e} ± 13.4	336.0 ^{e} ± 4.9	0.172 ^{d} ± 0.0	63.9 ^{def} ± 0.1
AM	2005	K	1876.5 ^{g} ± 11.7	268.0 ^{g} ± 8.5	0.164 ^{e} ± 0.0	62.8 ^{ef} ± 1.8
CB	2005	D	818.1 ^{j} ± 11.1	103.1 ^{k} ± 4.2	0.164 ^{e} ± 0.0	62.3 ^{ef} ± 1.1
VG	2005	D	2673.0 ^{b} ± 19.0	458.5 ^{a} ± 14.1	0.174 ^{cd} ± 0.0	77.8 ^{a} ± 1.1
ML	2005	D	2735.5 ^{a} ± 14.0	502.5 ^{a} ± 7.1	0.181 ^{c} ± 1.1	65.9 ^{cde} ± 0.1
XN	2006	K	1103.0 ^{i} ± 10.8	198.3 ^{i} ± 2.8	0.189 ^{b} ± 0.0	67.5 ^{c} ± 0.0
FT	2006	K	2516.0 ^{c} ± 9.9	398.5 ^{d} ± 8.5	0.172 ^{d} ± 0.0	61.5 ^{f} ± 1.4
AM	2006	K	2007.5 ^{d} ± 3.5	280.0 ^{f} ± 2.8	0.173 ^{d} ± 0.0	62.0 ^{f} ± 0.7
CB	2006	D	808.3 ^{j} ± 11.0	143.6 ^{j} ± 2.1	0.161 ^{e} ± 0.3	62.0 ^{f} ± 0.6
VG	2006	D	2571.5 ^{f} ± 6.4	415.5 ^{c} ± 4.0	0.198 ^{a} ± 0.0	79.6 ^{a} ± 0.1
ML	2006	D	2735.5 ^{a} ± 20.5	505.5 ^{a} ± 1.4	0.181 ^{c} ± 0.0	65.9 ^{cd} ± 1.3
Cultivar						
XN	05/06	K	1231.5 ^{b} ±181.7	212.2 ^{c} ± 116.1	0.188 ^{a} ± 0.0	67.2 ^{b} ± 0.4
FT	05/06	K	2389.8 ^{a} ±178.5	367.3 ^{b} ± 145.9	0.172 ^{abc} ± 0.0	62.7 ^{b} ± 1.6
AM	05/06	K	1942.0 ^{a} ±92.6	274.0 ^{c} ± 297.3	0.169 ^{bc} ± 0.0	62.4 ^{b} ± 1.5
CB	05/06	D	813.2 ^{b} ± 6.9	123.3 ^{d} ± 347.3	0.162 ^{c} ± 0.2	66.4 ^{b} ± 1.3
VG	05/06	D	2622.3 ^{a} ± 71.8	433.7 ^{ab} ± 8.5	0.186 ^{ab} ± 0.0	78.7 ^{a} ± 4.8
ML	05/06	D	2735.5 ^{a} ± 0.0	501.6 ^{a} ± 52.1	0.180 ^{abc} ± 0.8	65.4 ^{b} ± 0.9
SPI			3315.7 ± 148.7	614.2 ± 0.7	0.193 ± 0.0	67.5 ± 16.0
PPI			354.3 ± 37.1	74.1 ± 2.4	0.228 ± 0.0	81.8 ± 9.0
Biotype						
		Kabuli	1894.0 ^{a} ±582.0	286.9 ^{b} ± 540.0	0.179 ^{a} ± 0.0	64.1 ^{b} ± 2.5
		Desi	1973.6 ^{a} ± 933.0	355.4 ^{a} ± 891.4	0.176 ^{a} ± 0.5	70.2 ^{a} ± 6.8
Year						
		2005	1954.4 ^{a} ±757.9	315.7 ^{a} ± 737.6	0.174 ^{a} ± 0.5	66.4 ^{a} ± 5.5
		2006	1957.0 ^{a} ±797.7	322.8 ^{a} ± 738.6	0.178 ^{a} ± 0.1	67.8 ^{a} ± 6.4

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi , *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles, SPI = soy protein isolates, PPI = pea protein isolates

Though it is not plotted, the loss modulus (G″) and tan δ were monitored during the testing and are presented in Table 4.1.17. SPI had higher G' and G″ values than did the chickpea isolates, whereas values for PPI were lower than those for the chickpea isolates indicating chickpea protein formed gels that were less elastic and viscous than those for SPI, but greater than those for PPI.

4.1.2.4.3 Effect of NaCl concentration on heat-induced chickpea protein gels

Sodium chloride is extensively used as an additive along with plant protein (e.g. soy protein) to enhance emulsification, gelation and water and lipid retention in meat products, allowing the desired texture to be achieved (McMindes, 1991). The effect of NaCl concentration on G′, G″ and loss delta (δ) of 12 % (w/w) chickpea protein gelation is shown in Figure 4.1.11. The results indicate that NaCl exhibited a marked influence on chickpea isolate gel development during heating and cooling, and also affected the final gel properties.

Generally, Desi-type isolates had higher G′and G″ values than did those for Kabuli in the presence of NaCl. The optimum NaCl concentration for gel formation was 1% (0.25 M). In addition, there was a gradual decrease in both G' and G" and an increase in δ at NaCl concentrations of 2 and 3%. This could be due to higher protein solubilization of chickpea protein isolates by the salt solution, thereby creating an effective overlapping of the functional groups between adjacent protein molecules, a condition necessary for a network or gel formation (Catsimpoolas & Meyer, 1970). Such a NaCl concentration (0.2 M) was also found to promote optimum gel network characteristics for fababean (Arntfield et al., 1990b) and for red bean (Meng & Ma, 2002). At high salt concentrations (2% and 3%), the gel moduli were reduced. It has been suggested by Catsimpoolas & Meyer (1971) that hydrogen and ionic bonds are responsible for the stabilization of plant protein gels, and that addition of NaCl will decrease the viscosity of the gel if the concentration of NaCl is high enough to neutralize the charges stabilizing

the gel. Hence, usage of chickpea protein with NaCl at 1% in a food system would form a good quality gel.

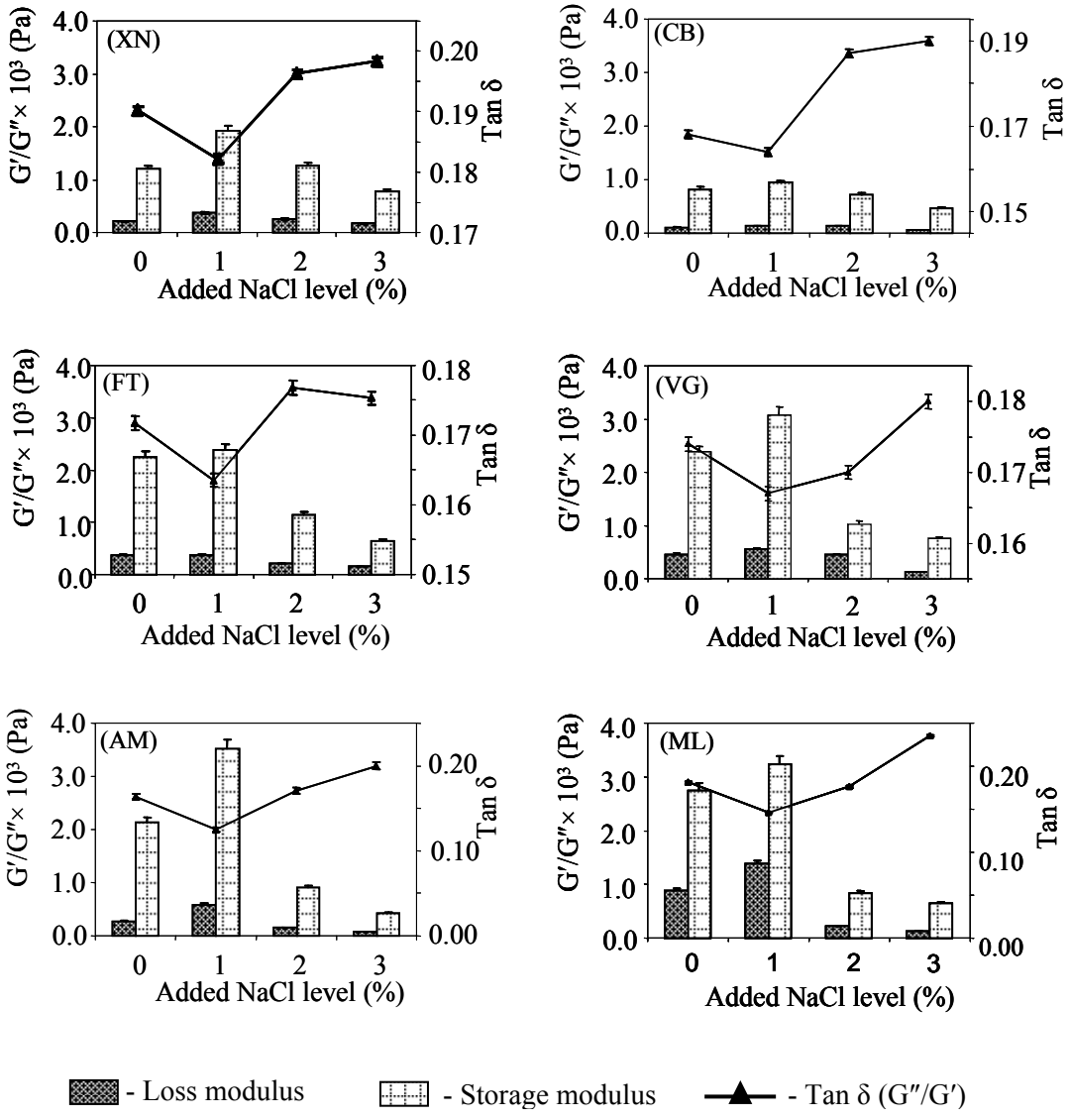

Figure 4.1.11 G′, G″ modulus and tan delta (δ)of 12% chickpea protein isolate dispersion in deionized water at pH 7.0 as a function of NaCl after heating and cooling cycle: XN = CDC Xena; FT = CDC Frontier; AM = Amit; CB = CDC Cabri; VG = CDC Vanguard; ML = Myles from 2005 harvest. Heating and cooling rate was 1 K/min.

4.1.2.4.4 Thermal stability of chickpea proteins

Differential scanning calorimetry (DSC) was performed on chickpea protein isolates to investigate their thermal stability. As shown in Figure 4.1.12, all showed a characteristic endotherm. The onset temperature (T_o), peak denaturation temperature (T_p), conclusion temperature (T_C), heat of transition or enthalpy (ΔH) and denaturation range (R) of chickpea protein isolates are shown in Table 4.1.18. The T_p for all protein isolates was less than 100°C. Chickpea proteins had a very broad peak with the T_p ranging from 87.8 to 94.2°C. There were no differences in thermal propeties between isolates from Kabuli and Desi chickpea, but they showed a significant year effect ($P<0.05$) in T_o and T_p. Kaur & Singh (2006) reported higher T_p values, ranging from 98.5-99.8°C, for Indian chickpea cultivars. Legume seed globulins have been found to possess T_p in the range of 83.8-107.8°C (Gorinstein et al., 1996).

The transition heat (ΔH) is used to monitor the proportion of the protein that denatures during the thermal process (Arntfield and Murray, 1981). For chickpea protein isolates, ΔH ranged from 2.6-4.1 J/g on a protein basis, which is accordance with a previously published value of 3.9 J/g for chickpea (84.8% protein, Paredes-Lopez et al., 1991).

Purified soy glycinin had only one major peak at 92.4°C, which is due to denaturation of their 11S globulin (Sheard, Ledward, & Mitchell, 1987; Horax et al., 2004; Zhong & Sun, 2000). However, a DSC thermogram of isolated β-conglycinin from soy (Figure 4.1.12) had two endothermic transitions which occurred at 74.2 and 92.8°C. The presence of a major peak at the lower temperature may be due to denaturation of the 7S globulin (Horax et al., 2004; Nagano et al., 1992; Sheard et al., 1987; Zhong & Sun, 2000) and at the higher temperature may represent denaturation of a cross-contaminating 11S component. This result further confirms the SDS-PAGE patterns of β-conglycinin isolates. Similar endothermic observations were reported for 11S contamination in 7S (Zhong & Sun, 2000).

As expected, SPI showed two peaks corresponding to denaturation of 7S and 11S proteins. The high enthalpy of the 11S peak (2.57 J/g) in SPI compared to the 7S peak (0.51 J/g) might indicate a higher amount of 11S globulin in the SPI than of the 7S globulin. Similar results were observed by Horax et al. (2004) for SPI. Pea protein isolate had only one major peak at 82.5°C, which may be due to the denaturation of a 7S component. Likewise, Horax, Hettiarchchy, Chen, & Jalaluddin (2004) studied cowpea protein isolates and reported one major peak, with a peak temperature ranging from 85.2–88.4°C, which they assigned to the denaturation of 7S globulins. Chickpea protein isolates denaturated at higher temperature (~80°C) compared to that observed for the of chickpea flour. Therefore, when chickpea proteins are going to be used in a food system, they shoud be cooked to at least 80°C to form a quality gel. As indicated above, besides differences in protein content and composition, interactions of proteins with residual salts in the isolates may have some effect on protein thermal stability (Murray, Arntfield, & Ismond, 1985). Therefore, further DSC studies of chickpea protein isolates in the presence of NaCl are needed.

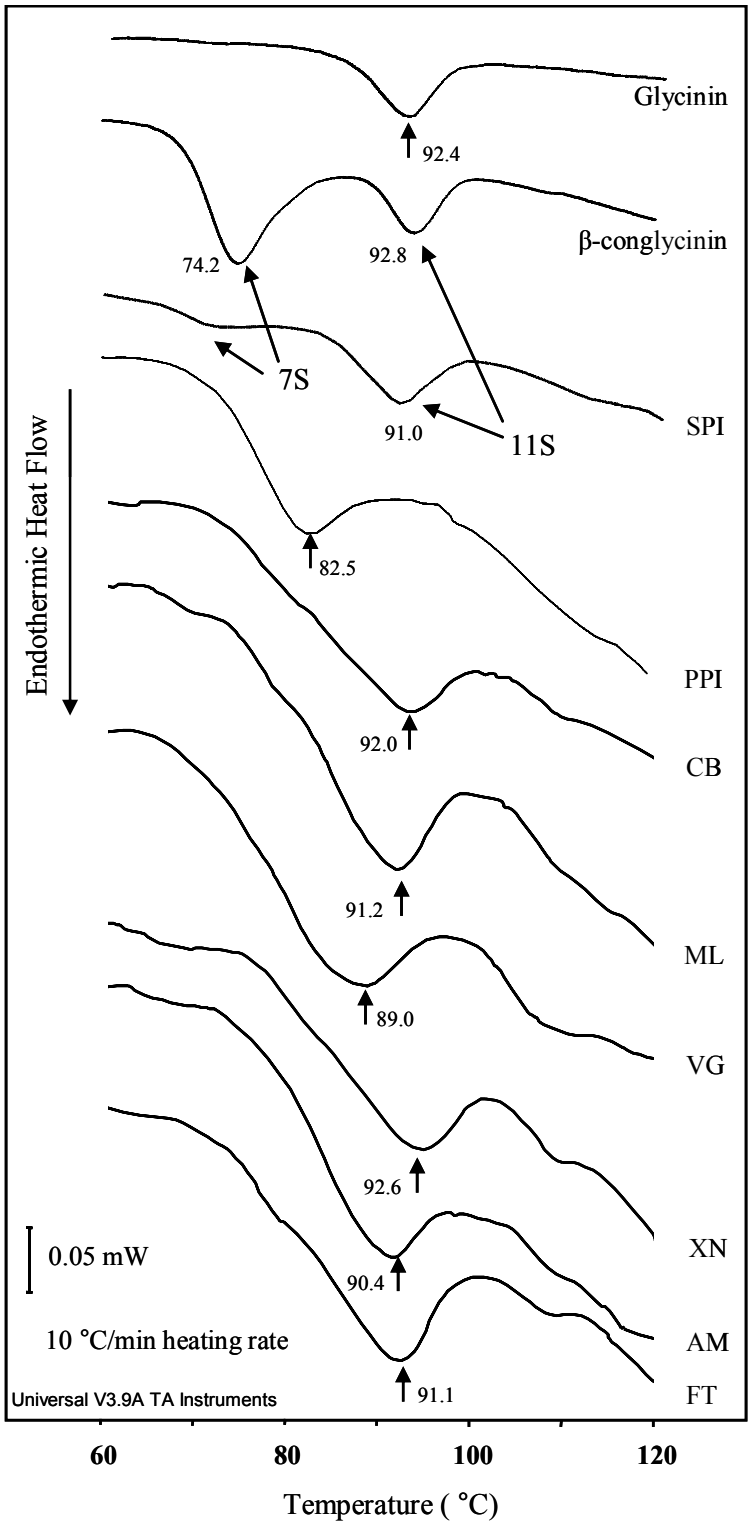

Figure 4.1.12 DSC thermograms of chickpea isolates at a 10% (w/v) protein concentration; heating rate: 10 °C/min. SPI = Soy protein isolates; PPI = Pea protein isolate; XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

Table 4.1.18 Thermal properties of protein isolates from different chickpea cultivars

Cultivar	Year	Biotype[1]	T_o [°C]	T_p [°C]	ΔH (J/g)
Year × cultivar					
XN	2005	K	79.6 ^{bc} ± 2.56	87.9 ^{ef} ± 0.85	3.0 ^{a} ± 0.38
FT	2005	K	78.2 ^{bc} ± 0.84	91.1 ^{cd} ± 0.14	3.9 ^{a} ± 0.64
AM	2005	K	79.8 ^{bc} ± 1.77	90.4 ^{cde} ± 1.04	2.8 ^{a} ± 0.82
CB	2005	D	76.2 ^{c} ± 0.60	92.0 ^{bc} ± 0.26	3.0 ^{a} ± 0.19
VG	2005	D	78.4 ^{bc} ± 0.91	89.0 ^{def} ± 0.83	4.0 ^{a} ± 0.81
ML	2005	D	80.1 ^{bc} ± 0.99	90.0 ^{cdef} ± 0.51	3.3 ^{a} ± 0.07
XN	2006	K	81.1 ^{bc}± 1.62	91.1 ^{cd} ± 0.16	4.0 ^{a} ± 0.01
FT	2006	K	82.1 ^{bc} ± 2.95	87.6 ^{f} ± 0.03	3.2 ^{a} ± 0.01
AM	2006	K	92.1 ^{a} ± 0.64	97.4 ^{a} ± 0.31	3.5 ^{a} ± 0.12
CB	2006	D	85.3 ^{b} ± 0.98	93.7 ^{b} ± 0.74	2.4 ^{a} ± 0.07
VG	2006	D	75.3 ^{c} ± 0.15	96.4 ^{a} ± 0.93	2.6 ^{a} ± 0.00
ML	2006	D	80.9 ^{bc} ± 1.32	90.0 ^{cdef} ± 0.13	3.3 ^{a} ± 0.57
Cultivar					
XN	05/06	K	80.2 ^{ab} ± 2.21	89.2 ^{a} ± 0.18	3.4 ^{a} ± 0.14
FT	05/06	K	80.0 ^{ab} ± 9.80	89.7 ^{a} ± 4.44	3.6 ^{a} ± 0.30
AM	05/06	K	84.7 ^{a} ± 3.94	93.2 ^{a} ± 2.35	3.1 ^{a} ± 0.27
CB	05/06	D	79.9 ^{ab} ± 0.63	92.7 ^{a} ± 3.09	2.8 ^{a} ± 0.29
VG	05/06	D	76.8 ^{b} ± 1.91	92.8 ^{a} ± 0.69	3.3 ^{a} ± 0.17
ML	05/06	D	79.9 ^{ab} ± 4.05	90.0 ^{a} ± 1.88	3.3 ^{a} ± 0.11
SPI			85.6 ± 1.47	91.1 ± 0.12	2.8 ± 0.00
PPI			73.6 ± 1.01	82.2 ± 0.27	1.6 ± 0.05
Biotype					
		Kabuli	81.7 ^{a} ± 5.15	91.9 ^{a} ± 3.70	3.4 ^{a} ± 0.52
		Desi	79.0 ^{a} ± 4.07	90.7 ^{a} ± 2.69	3.1 ^{a} ± 0.60
Year					
		2005	78.5 ^{b} ± 1.62	90.1 ^{b} ± 1.54	3.3 ^{a} ± 0.51
		2006	82.5 ^{a} ± 5.65	92.7 ^{a} ± 3.73	3.2 ^{a} ± 0.63

Mean (±SD) of triplicate analysis
Means followed by the same letter within column and section do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi
Var: XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles
T_o, onset temperature of denaturation, T_p, peak temperature, ΔH, enthalpy of denaturation

4.1.2.5 Functional properties

The functional properties of protein isolates primarily determine their utility in food products. The biotypes belonging to two distinct chickpea groups showed a large difference in certain functional properties.

4.1.2.5.1 Nitrogen solubility index (NSI)

Nitrogen solubility index was determined by the standard method of AACC (1995) 46-23. Minimum protein solubility of different chickpea cultivars was around pH 4.3, with over 90% of the proteins soluble at pH 6 or above (Figure 4.1.13). These observations are in agreement with those of Sánchez-Vioque et al. (1999), who observed an isoelectric pH of 4.3 for chickpea protein isolates. Such patterns were also reported for other legumes, such as Chinese indigenous legume (Chau et al., 1997), winged bean (Sathe et al., 1982b), lupin (Sathe et al., 1982a) and cowpea and pea vicilin (Rangel et al., 2003). Soy protein isolate and PPI had minimum protein solubilities at pH 4.5, and at pH 6 only about 80% of the nitrogen was soluble. CDC Xena, PPI and SPI exhibited a markedly higher insolubility over a broader range of pH (CDC Xena: 3-5 and SPI/PPI: 4-6) than did the other chickpea protein isolates (4.0-5.0). High protein solubility in both acidic and basic pH regions could be an important characteristic in food formulations.

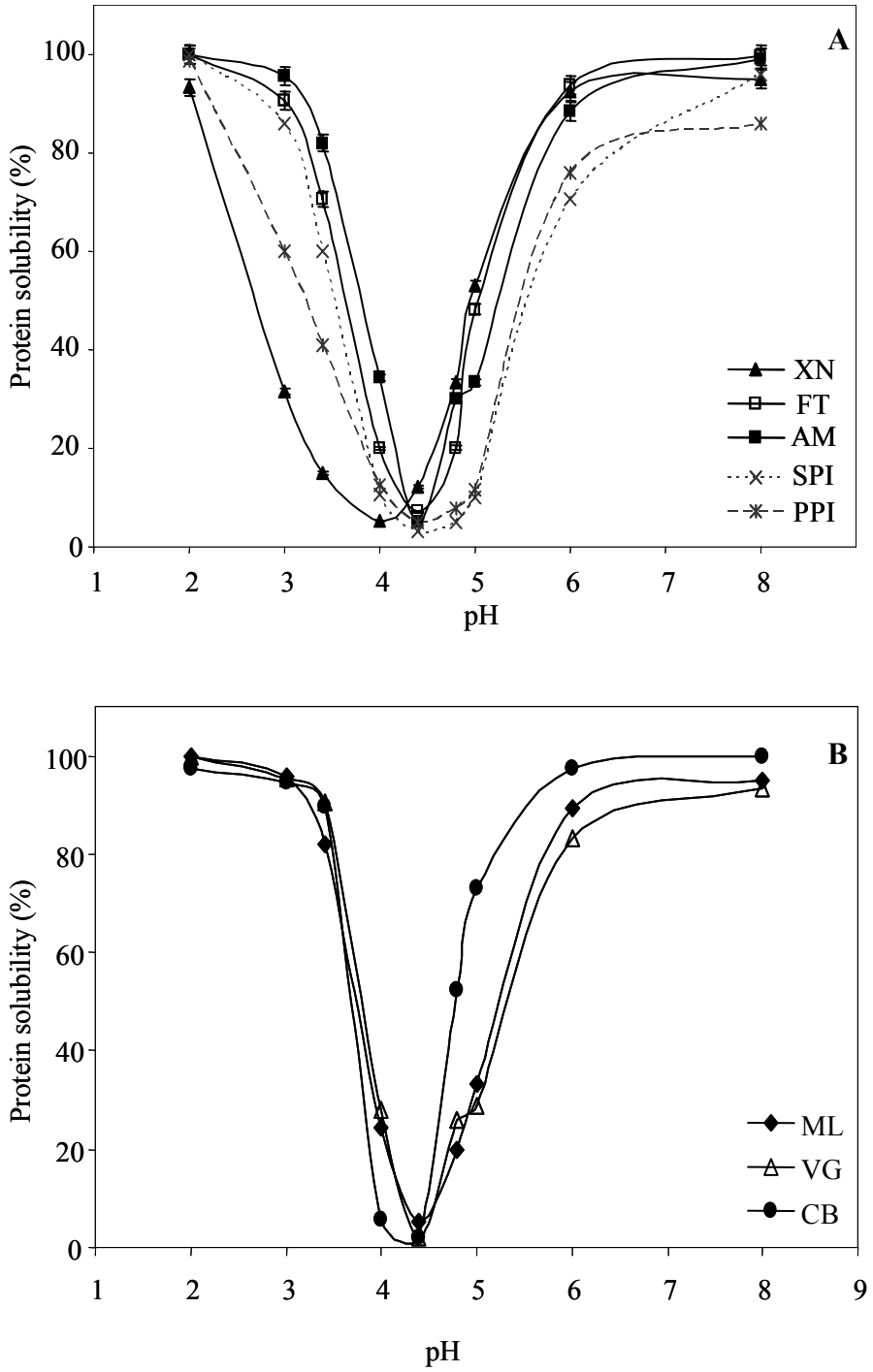

Figure 4.1.13 Solubility curves of the chickpea protein as a function of pH: (A) Kabuli var. (B) Desi var. SPI = Soy protein isolates; PP = Pea protein isolates; XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

4.1.2.5.2 Water holding capacity (WHC) and oil absorption capacity (OAC)

WHC of different chickpea isolates showed both 'biotype' and 'year' effects ($P<0.05$) (Table 4.1.19). The WHCs of Kabuli-type isolates were significantly ($P<0.05$) higher than those of the Desi-type. The mean WHC value of chickpea protein isolates ranged from 2.34 to 4.31 g/g. CDC Xena, Myles and SPI had the highest WHCs ($P<0.05$), whereas, the lowest WHC was observed for CDC Cabri. The reason why CDC Cabri had the lowest WHC may be related to its low protein content when compared with the other protein isolates. The WHC of PPI was lower than the WHC of chickpea and SPI. Moreover, WHC of protein isolates in the present study compared favorably to isolates from Great Northern bean (2.73 g/g, Sathe and Salunkhe, 1981), winged bean (3.51 g/g, Sathe et al., 1982) and Indian chickpea cultivars (2.4-3.4 g/g, Kaur and Singh, 2005). Basically, these differences in WHC might be attributed to different protein conformations and variations in the number and nature of water-binding sites on protein molecules (Chou & Morr, 1979). The protein isolates from different chickpea cultivars exhibited significantly different WHC values for the two harvest years (Figure 4.1.14). Furthermore, the protein isolates from different chickpea cultivars exhibited significantly higher WHC as compared to their flours (see Table 4.1.10).

The oil absorption capacity (OAC) of proteins is also important as it improves mouth feel and retains flavour in a protein matrix (Kinsella, 1981). The OACs of Kabuli-type isolates were significantly ($P<0.05$) higher than those of the Desi-type. OAC of different chickpea protein isolates ranged from 3.06 to 5.74 g/g (Table 4.1.19). Interestingly, CDC Xena (Kabuli-type) and Myles (Desi-type) had superior OACs to SPI and PPI. The higher OAC might be attributed to a higher level of nonpolar side chains in their protein molecules (Chau et al., 1997). The high WHC and OAC of chickpea proteins may be useful in food applications such as emulsion-type products. Generally, oil absorption capacity of chickpea protein isolates was higher than that of chickpea flour. Proteins that are more hydrophobic show superior binding

of lipids (Horax et al., 2004), implying that non-polar amino acid side chains bound the corn oil more than that of the flour.

Table 4.1.19 Water holding and oil absorption capacity of chickpea proteins, SPI and PPI

Var.	Year	Biotype	WHC (g/g)	OAC (g/g)
Cultivar				
XN	05/06	K	4.28 [a] ± 0.36	5.74 [a] ± 0.36
FT	05/06	K	3.46 [b] ± 0.43	3.90 [c] ± 0.38
AM	05/06	K	3.28 [b] ± 0.97	3.20 [d] ± 1.07
CB	05/06	D	2.34 [c] ± 0.18	3.35 [cd] ± 0.23
VG	05/06	D	3.28 [b] ± 0.71	3.06 [d] ± 0.18
ML	05/06	D	4.31 [a] ± 0.32	4.83 [b] ± 0.17
SPI			4.27 ± 0.28	4.16 ± 0.04
PPI			2.68 ± 0.27	2.76 ± 0.11
Biotype				
		Kabuli	3.68 [a] ± 0.67	4.28 [a] ± 1.22
		Desi	3.31 [b] ± 0.92	3.75 [b] ± 0.83
Year				
		2005	3.71 [a] ± 0.79	4.15 [a] ± 0.92
		2006	3.27 [b] ± 0.82	3.88 [a] ± 1.21

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi ; *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

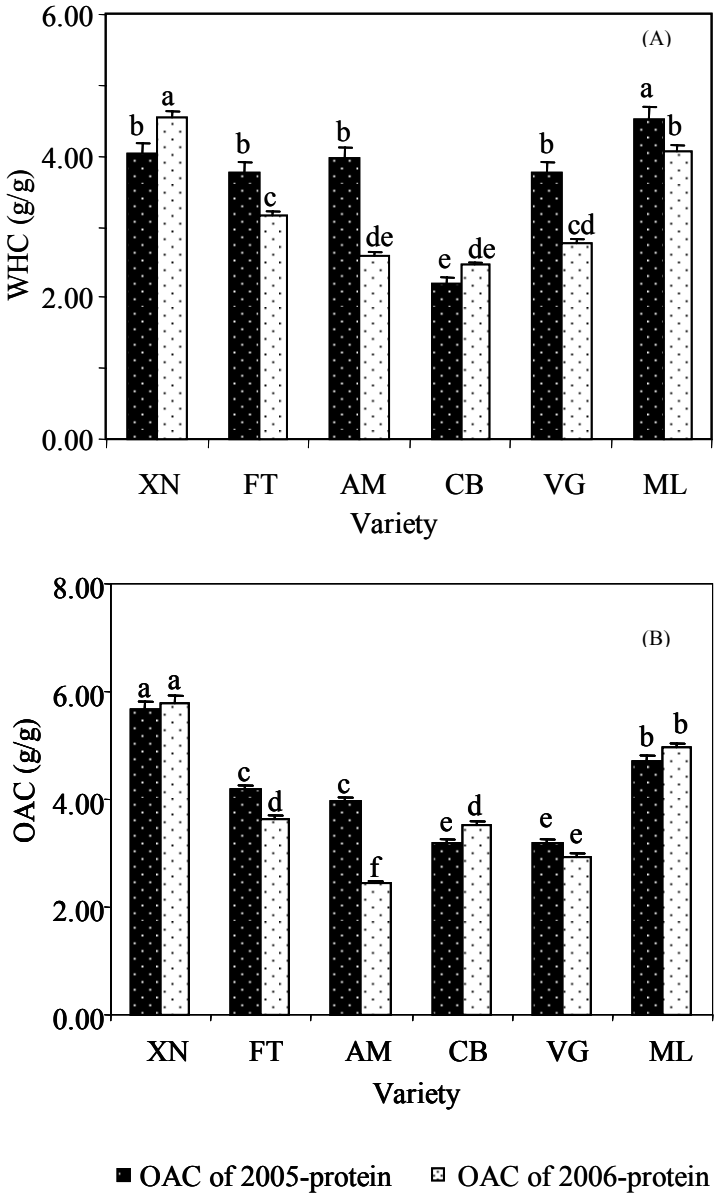

Figure 4.1.14 (A) Water holding capacity (WHC) (B) Oil absorption capacity (OAC) of flour and protein isolates from chickpea cultivars; XN: CDC Xena, FT: CDC Frontier, AM: Amit, CB: CDC Cabri, VG: CDC Vanguard, ML: Myles.

4.1.2.5.3 Emulsion ability index (EAI) and emulsion stability index (ESI)

Emulsion ability is defined as the ability of the protein to emulsify oil. The protein isolates from both Kabuli and Desi showed higher EAI and ESI than did the chickpea flours. For EAI, protein isolates from Kabuli cultivars showed higher values (1.13-1.30) than did Desi cultivars (0.88-1.15). Chickpea isolates from various chickpea cultivars (except CDC Vanguard and Myles) were similar in their ability to emulsify corn oil (Table 4.1.20), and similar to SPI.

Table 4.1.20 Cultivar, biotype and year effects on EAI and ESI of various protein isolates

Var.	Year	Biotype[1]	EAI	ESI
Cultivar				
XN	05/06	K	1.30 [a] ± 0.00	26.65 [a] ± 0.21
FT	05/06	K	1.13 [ab] ± 0.04	25.09 [a] ± 0.15
AM	05/06	K	1.30 [a] ± 0.01	20.30 [bc] ± 0.71
CB	05/06	D	1.15 [ab] ± 0.21	19.25 [c] ± 0.49
VG	05/06	D	0.94 [bc] ± 0.06	21.15 [b] ± 0.21
ML	05/06	D	0.88 [c] ± 0.04	21.35 [b] ± 1.20
SPI			1.28 ± 0.04	29.29 ± 0.22
PPI			0.89 ± 0.02	18.31 ± 0.08
Biotype (chickpea)				
		Kabuli	1.24 [a] ± 0.09	24.01 [a] ± 2.98
		Desi	0.99 [b] ± 0.16	20.58 [b] ± 1.19
Year				
		2005	1.08 [a] ± 0.44	22.27 [a] ± 8.81
		2006	1.15 [a] ± 0.47	22.33 [a] ± 8.88

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi ; *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles; SPI = native soy protein isolates; PPI = native pea protein isolates

Similarly chickpea proteins from Kabuli-types had significantly higher ESI (20.30-26.65) than did protein from Desi-types (19.25-21.35) (Table 4.1.20). The higher ESI of Kabuli isolate might be attributed to a higher level of nonpolar side chains in their protein molecules (Chau et al., 1997) that might have stabilized the emulsion better. No significant differences ($P<0.05$) were observed due to production year for EAI and ESI of chickpea protein isolates. SPI had the highest ESI and PPI and CDC Cabri had the lowest values. The emulsifying activity of cowpea proteins and soy proteins has been reported to range from 0.59 to 0.72 and from 1.10 to 1.21, respectively (Horax et al., 2004). The high emulsifying activity of legume proteins may be because the surface of legume protein contains a high number of hydrophobic residues, which can disperse the droplets of oil in the aqueous continuous phase of the solution.

The turbidity of the emulsion was plotted (Figure 4.1.15) as the ordinate and standing time after emulsion formation as the abscissa according to the method of Pearce & Kinsella (1978). The decrease of absorbance followed first order kinetics (Pearce & Kinsella, 1978). Chickpea curves were in between the SPI (above) and PPI curves (below). These results indicate that the chickpea protein isolates investigated (especially from Kabuli) were effective emulsifiers, making them potentially useful in applications such as the manufacture of mayonnaise, sausages, and seasonings.

Correlation analyses of chickpea protein isolates, shown in Table 4.1.21, showed a significant and positive correlation of protein content with WHC ($r = 0.71$, $P<0.01$), OAC ($r = 0.55$, $P<0.05$) and ESI ($r = 0.66$, $P<0.05$). There were also significant negative relationships between protein and crude fat content ($r = -0.70$, $P<0.01$) and total lipid content ($r = -0.60$, $P<0.05$) in the chickpea protein isolates. Other important positive relationships between WHC and storage modulus and loss modulus ($r = 0.56$ and 0.66, respectively, $P<0.05$) were observed. Finally, among functional properties, WHC and OAC were significantly and positively correlated with the emulsion stability of chickpea proteins.

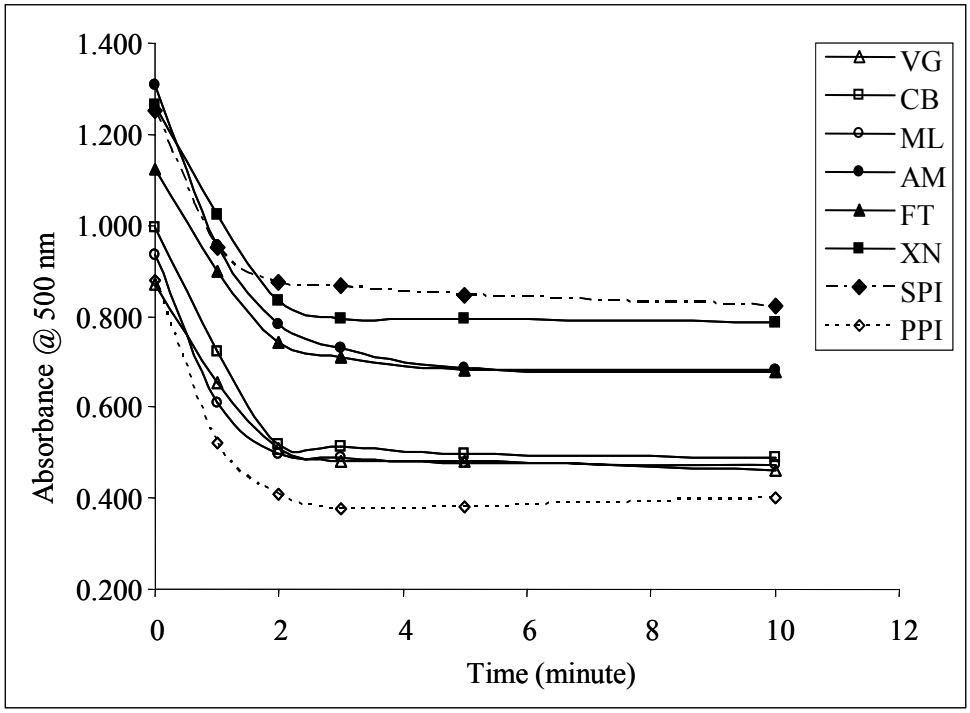

Figure 4.1.15 Time development of the emulsion system from different chickpea cultivars (A) Flour (B) protein isolates from 2005 harvesting year (λ_{max} : 500 nm). *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles; SPI = native soy protein isolates; PPI = native pea protein isolates

Table 4.1.21 Correlation coefficients (*r*) of chemical, thermal and functional properties chickpea protein isolates (combined data, n=12)

	2	3	4	5	6	7	8	9
1. Protein content	-0.04	-0.70**	-0.60*	0.28	0.19	-0.21	0.31	0.23
2. Ash	1	-0.53*	-0.46*	0.71**	-0.21	-0.65**	-0.28	-0.48
3. Crude fat		1	0.79***	-0.72**	-0.05	0.45	0.66**	-0.07
4. Total fat			1	-0.62**	-0.01	0.21	0.61*	0.00
5. Total starch				1	-0.15	-0.49	-0.32	-0.22
6. T_o (onset)					1	0.58*	-0.18	0.41
7. T_p (peak)						1	0.00	0.40
8. ΔH							1	-0.19
9. G'								1
10. G"								
11. δ								
12. Onset								
13.WHC								
14. OAC								
15. EAI								
16. ESI								

*, **, *** = Significant at P<0.05, 0.01 and 0.001, respectively.
1 – 5 proximate values; 6 – 8 DSC parameters; 9-11 –Rheology data ; 13 – water holding capacity; 14 – oil absorption capacity; 15-16 emulsion properties.

Table 4.1.21 continued …

	10	11	12	13	14	15	16
1. Protein content	0.35	0.34	-0.01	0.71**	0.55*	0.29	0.66**
2. Ash	-0.41	0.50	0.32	-0.44	-0.38	-0.28	-0.31
3. Crude fat	-0.19	-0.62**	-0.27	-0.20	0.01	-0.04	-0.34
4. Total fat	-0.18	-0.50*	-0.29	-0.00	0.07	-0.11	-0.39
5. Total starch	-0.14	0.62*	0.35	-0.19	-0.35	-0.27	-0.20
6. T_o (onset)	0.38	0.46	-0.56*	0.15	0.08	0.69**	0.36
7. T_p (peak)	0.31	-0.58*	-0.42	-0.04	-0.03	0.42	0.12
8. ΔH	-0.27	-0.24	-0.13	0.04	0.15	-0.24	-0.26
9. G'	0.98***	-0.27	-0.32	0.56*	0.22	0.07	0.56*
10. G"	1	-0.18	-0.24	0.62*	0.27	0.02	-0.35
11. δ		1	0.65**	-0.07	-0.17	-0.34	-0.06
12. Onset			1	-0.29	-0.40	0.51*	0.59*
13.WHC				1	0.80***	0.19	0.66**
14. OAC					1	0.28	0.62*
15. EAI						1	0.53*
16. ESI							1

*, **, *** = Significant at P<0.05, 0.01 and 0.001, respectively.
1 – 5 proximate values; 6 – 8 DSC parameters; 9-11 –Rheology data ; 13 – water holding capacity; 14 – oil absorption capacity; 15-16 emulsion properties.

4.1.2.6 Summary and conclusions

Protein isolates from Desi and Kabuli chickpeas were prepared by wet extraction. Proximate composition (moisture, protein, ash, and fat), functional (solubility index, water holding capacity, oil absorption capacity, emulsifying ability index and emulsion stability index) and thermal characteristics (critical gelation concentration, rheological and denaturation properties) of these protein isolates were investigated and compared with soy protein and pea protein isolates. Chickpea protein isolates had 73.0-82.0% protein, 3.1-4.2% ash and 7.1-9.8% fat (dry weight basis). Protein isolates from CDC Xena, among Kabuli-types and Myles, among Desi-types, as well as SPIs had the highest water holding and oil absorption capacities.

Isolates from Kabuli cultivars exhibited higher emulsifying ability indices and emulsion stability indeces than did Desi isolates. The SPI had an emulsion stability index as high as the Kabuli isolates and it gave the most stable emulsion. Peak temperatures of the endothermic peaks for chickpea protein isolates ranged from 89.0 to 92.0°C, and the enthalpy of denaturation was 2.4-4.0 J/g. Protein solubility curves showed that minimum solubility ranged between pH 4.1-4.4. Furthermore, solubility characteristics showed over 90% solubility at pH 6 for all chickpea cultivars, but only near 80% for pea and soy protein isolates at pH 6. The heat-induced gelation properties of Kabuli chickpea isolates and Desi chickpea isolates showed a minimum protein concentration required form a gel structure ranging from 8 to 11% (w/v, protein basis). The onset temperature for structure development (small amplitude oscillatory testing) ranged from 61.5 to 78°C. Protein isolates from Kabuli chickpea generally had lower onset temperatures than did those for Desi. Most functional properties compared favorably to the soy isolate, and better than those of the pea protein isolates. Both Kabuli and Desi protein isolates with attractive physico-chemical, thermal and functional properties, showed potential for food use.

4.1.3 Proximate and thermal properties of starches from different chickpea cultivars.

4.1.3.1 Chemical composition of chickpea starch

The chemical characteristics of isolated chickpea starches are summarized in Table 4.1.22. From 1 kg of chickpea seeds, approximately 0.42 kg of starch was obtained, which is equivalent to a starch recovery of 42%. Generally, there was no biotype or year effect for the chemical composition of chickpea starch (0.46% protein, 0.04% fat, 0.20% ash, 34.1% amylose, 95.2% total starch). There were not significant correlations among chemical properties of chickpea starches from different cultivars of both years (data not shown). The purity of chickpea starch was above 93.0%. The isolated starches were characterized by low protein and low fat contents of 0.44-0.48% and 0.01-0.07%, respectively. The ash contents, reflecting contamination by fine fibre, of various chickpea starches ranged from 0.14 to 0.35%. These values are in accordance with those reported earlier for chickpea starches (Hoover & Ratnayake, 2002). Chickpea starches had amylose contents ranging from 31.0-36.8% (Table 4.1.22). The highest amylose content was observed for CDC Xena (36.81 %), and the lowest for CDC Frontier (31.04 %).

Legume starches are characterized by an intermediate amylose content of 24-65% (Hoover & Ratnayake, 2002). For example, the amylose content was 30.2-34.6% in black gram starch (Singh et al., 2004), 32.7% in baby lima bean starch (Betancur, Chel, Rosa, Gloria, & Ortiz, 2001) and 27% in chickpea starch (Meares et al., 2004).

Table 4.1.22 Year × cultivar, cultivar and year effects on chemical composition of chickpea starches on dry weight basis

Var.	Year	Biotype [1]	Protein[2] %)	Fat (%)	Ash (%)
Year × cultivar					
XN	2005	K	0.47 [a] ± 0.04	0.06 [cb] ± 0.00	0.22 [c] ± 0.01
FT	2005	K	0.47 [a] ± 0.02	0.06 [b] ± 0.00	0.16 [de] ± 0.01
AM	2005	K	0.47 [a] ± 0.05	0.03 [e] ± 0.00	0.15 [def] ± 0.02
CB	2005	D	0.46 [a] ± 0.01	0.01 [f] ± 0.00	0.11 [f] ± 0.03
VG	2005	D	0.46 [a] ± 0.00	0.01 [f] ± 0.00	0.11 [f] ± 0.04
ML	2005	D	0.46 [a] ± 0.52	0.04 [d] ± 0.01	0.14 [def] ± 0.05
XN	2006	K	0.45 [a] ± 0.03	0.07 [a] ± 0.00	0.13 [ef] ± 0.01
FT	2006	K	0.45 [a] ± 0.02	0.05 [cbd] ± 0.00	0.28 [b] ± 0.03
AM	2006	K	0.48 [a] ± 0.04	0.06 [b] ± 0.00	0.55 [a] ± 0.01
CB	2006	D	0.49 [a] ± 0.05	0.02 [cd] ± 0.00	0.16 [de] ± 0.03
VG	2006	D	0.41 [b] ± 0.05	0.01 [f] ± 0.00	0.17 [de] ± 0.02
ML	2006	D	0.41 [b] ± 0.00	0.02 [f] ± 0.00	0.19 [cd] ± 0.00
Cultivar					
XN	05/06	K	0.46 [a] ± 0.01	0.07 [a] ± 0.01	0.18 [a] ±0.06
FT	05/06	K	0.46 [a] ± 0.01	0.06 [a] ± 0.01	0.22 [a] ± 0.08
AM	05/06	K	0.48 [a] ± 0.01	0.05 [a] ± 0.02	0.35 [a] ± 0.28
CB	05/06	D	0.48 [a] ± 0.02	0.02 [b] ± 0.01	0.14 [a] ± 0.04
VG	05/06	D	0.44 [a] ± 0.04	0.01 [b] ± 0.00	0.14 [a] ± 0.04
ML	05/06	D	0.44 [a] ± 0.04	0.03 [b] ± 0.01	0.17 [a] ± 0.04
Biotype					
		Kabuli	0.47 [a] ±0.01	0.06 [a] ± 0.01	0.24 [a] ± 0.16
		Desi	0.44 [a] ± 0.03	0.02 [b] ± 0.02	0.15 [a] ± 0.03
Year					
		2005	0.47 [a] ± 0.01	0.04 [a] ± 0.02	0.25 [a] ± 0.04
		2006	0.45 [a] ± 0.03	0.04 [a] ± 0.02	0.15 [a] ± 0.16

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi , [2]Total nitrogen × 6.25
Var: XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

Table 4.1.22 Continued …

Var.	Year	Biotype [1]	Amylose (%)	Total starch (%)
Year × cultivar				
XN	2005	K	36.49 [a] ± 0.01	97.59 [ab] ± 0.21
FT	2005	K	30.87 [d] ± 0.50	96.10 [b] ± 0.22
AM	2005	K	35.02 [b] ± 0.39	92.60 [c] ± 0.16
CB	2005	D	35.80 [ab] ± 0.50	97.13 [b] ± 1.55
VG	2005	D	32.16 [c] ± 0.22	99.69 [a] ± 0.19
ML	2005	D	32.65 [c] ± 0.58	96.60 [b] ± 0.17
XN	2006	K	37.13 [a] ± 0.01	98.44 [ab] ± 0.04
FT	2006	K	31.21 [c] ± 0.75	93.56 [c] ± 0.69
AM	2006	K	35.50 [ab] ± 0.39	97.80 [ab] ± 0.62
CB	2006	D	36.11 [a] ± 0.30	88.93 [d] ± 1.11
VG	2006	D	32.23 [c] ± 0.22	92.32 [c] ± 0.67
ML	2006	D	34.18 [b] ± 0.58	91.27 [c] ± 1.55
Cultivar				
XN	05/06	K	36.81 [a] ± 0.45	98.01 [a] ±0.60
FT	05/06	K	31.04 [d] ± 0.24	94.83 [a] ±1.80
AM	05/06	K	35.26 [b] ± 0.34	95.20 [a] ±3.68
CB	05/06	D	35.96 [ab] ± 0.23	93.03 [a] ± 5.80
VG	05/06	D	32.19 [cd] ± 0.05	96.00 [a] ±5.21
ML	05/06	D	33.41 [c] ± 1.08	93.94 [a] ±3.77
Biotype				
		Kabuli	34.37 [a] ± 2.42	96.02 [a] ±2.42
		Desi	33.85 [a] ± 4.11	94.32 [a] ±4.11
Year				
		2005	33.83 [a] ± 2.33	96.62 [a] ±2.33
		2006	34.39 [a] ± 3.74	93.72 [a] ±4.11

Means (±SD) of triplicate analysis per year and all data on dry weight basis
Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)
[1] Biotype: K = Kabuli, D = Desi ,*Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

4.1.3.2 Thermal properties

4.1.3.2.1 Pasting properties of chickpea starches

Pasting temperature provides as indication of the minimum temperature that is required when cooking starches. Furthermore, starch paste behavior in aqueous systems depends on the physical and chemical characteristics of the starch granules, such as mean granule size, granule size distribution, amylose/amlopectin ratio and mineral content (Madsen & Christensen, 1996). The results from the rapid viscoanalysis (RVA) of chickpea starches are summarized in Table 4.1.23. Overall this analysis demonstrated that biotype had a large influence. Kabuli-type starches had higher pasting characteristics than did the Desi-types starches, with the exception of pasting temperature. Pasting temperatures of starches from different chickpea cultivars ranged between 70.2 and 74.8°C. This might be due to the granule composition, amylose/amylopectin composition and other components in the starch such as proteins, lipids, sugars (Thomas & Atwell, 1997) as well as plant growth conditions (Tester et al., 1997). Within cultivars, the highest pasting temperature was observed for Myles starch, and the lowest for CDC Frontier starch. Moreover, pasting temperatures of isolated starches were higher than those of the flours (see Table 4.1.9).

Singh et al. (2004) reported pasting temperatures of starches from chickpea cultivars ranging between 75.1-77.1°C. Lineback and Ke (1975) reported pasting temperature of 68.5°C for chickpea starch and 67.0°C for horse bean starch. Pasting temperatures of 66.0°C for faba-bean starch and 71°C for mung bean starch have been reported by Naivikul & D'Appolonia (1979). Singh et al. (2004) reported pasting temperature between 75.8 and 80.3°C for black gram starches.

Table 4.1.23 Year × cultivar, cultivar and year effects on pasting properties of starches from different chickpea cultivars

Var.	Year	Genotype	peak1 (RVU)	trough1 (RVU)	Breakdown (RVU)	Final viscosity (RVU)	Setback (RVU)	Pasting temperature (°C)
Year × cultivar								
XN	2005	K	158.3 ^{a} ± 1.7	107.6 ^{c} ± 2.8	51.0 ^{a} ± 2.5	293.1 ^{ab} ± 1.0	185.5 ^{a} ± 2.8	71.9 ^{cd} ± 1.10
FT	2005	K	140.7 ^{b} ± 2.7	93.9 ^{def} ± 2.4	41.8 ^{ab} ± 0.3	265.6 ^{bc} ± 1.2	171.7 ^{a} ± 4.8	70.5 ^{e} ± 0.35
AM	2005	K	159.4 ^{a} ± 0.1	121.2 ^{b} ± 3.3	38.2 ^{bc} ± 2.2	310.6 ^{ef} ± 0.5	189.4 ^{a} ± 4.8	71.2 ^{ed} ± 0.14
CB	2005	D	119.3 ^{d} ± 0.6	81.5 ^{g} ± 0.1	37.8 ^{bc} ± 0.0	222.0 ^{f} ± 1.5	140.5 ^{bc} ± 1.4	72.8 ^{bc} ± 0.71
VG	2005	D	128.2 ^{c} ± 3.5	97.8 ^{de} ± 1.1	30.3 ^{c} ± 2.4	248.5 ^{de} ± 3.9	150.6 ^{bc} ± 2.8	73.4 ^{b} ± 0.35
ML	2005	D	91.2 ^{f} ± 1.6	85.8 ^{gf} ± 3.3	6.0 ^{d} ± 1.8	180.5 ^{g} ± 1.9	94.7 ^{ed} ± 1.3	75.2 ^{a} ± 0.64
XN	2006	K	164.7 ^{a} ± 3.6	132.2 ^{a} ± 4.7	32.9 ^{c} ± 1.7	290.0 ^{ab} ± 6.8	158.1 ^{b} ± 4.0	72.0 ^{cd} ± 0.05
FT	2006	K	162.4 ^{a} ± 4.7	127.3 ^{ab} ± 1.6	35.1 ^{bc} ± 8.4	259.1 ^{dc} ± 4.6	131.8 ^{c} ± 8.6	69.7 ^{f} ± 0.22
AM	2006	K	159.1 ^{a} ± 2.7	121.2 ^{b} ± 0.8	37.9 ^{bc} ± 3.3	233.8 ^{c} ± 7.3	112.6 ^{d} ± 7.9	71.9 ^{cd} ± 0.50
CB	2006	D	101.1 ^{e} ± 5.8	100.8 ^{cd} ± 5.2	36.6 ^{a} ± 0.9	158.7 ^{g} ± 5.4	57.8 ^{f} ± 6.4	72.8 ^{c} ± 0.30
VG	2006	D	116.6 ^{d} ± 6.6	89.3 ^{gef} ± 5.8	27.3 ^{c} ± 5.2	167.0 ^{g} ± 7.1	77.7 ^{e} ± 4.5	74.6 ^{a} ± 0.26
ML	2006	D	91.2 ^{f} ± 1.6	85.8 ^{gf} ± 3.3	5.4 ^{d} ± 0.7	180.5 ^{g} ± 1.9	94.7 ^{ed} ± 1.3	74.5 ^{a} ±0.50
Cultivar								
XN	05/06	K	160.9 ^{a} ± 4.4	117.4 ^{ab} ± 14.4	43.6 ^{a} ±10.4	291.8 ^{a} ± 7.5	174.5 ^{a} ± 15.4	72.0 ^{dc} ± 0.55
FT	05/06	K	151.0 ^{a} ± 11.1	107.4 ^{b} ± 18.3	41.6 ^{a} ± 8.4	269.0 ^{a} ± 17.4	161.6 ^{a} ± 30.5	70.2 ^{e} ± 0.52
AM	05/06	K	159.3 ^{a} ± 1.9	121.2 ^{a} ± 2.7	38.1 ^{a} ± 3.5	279.9 ^{a} ± 42.4	158.7 ^{a} ± 42.5	71.5 ^{d} ± 0.42
CB	05/06	D	112.0 ^{c} ± 10.8	89.3 ^{c} ± 11.2	37.3 ^{a} ± 0.9	196.7 ^{b} ± 35.6	121.5 ^{b} ± 45.5	72.6 ^{c} ± 0.44
VG	05/06	D	123.5 ^{b} ± 7.1	94.4 ^{c} ± 6.1	29.1 ^{b} ± 4.3	215.9 ^{b} ± 41.8	107.4 ^{b} ± 37.4	73.9 ^{b} ± 0.50
ML	05/06	D	91.2 ^{d} ± 1.4	85.8 ^{c} ± 2.9	5.7 ^{c} ± 1.3	180.5 ^{b} ± 1.7	94.7 ^{b} ± 1.2	74.8 ^{a} ± 0.57
Biotype								
		Kabuli	157.0 ^{a} ± 7.8	115.3 ^{a} ± 13.9	41.1 ^{a} ± 7.7	280.2 ^{a} ± 26.6	165.0 ^{a} ± 30.0	71.2 ^{b} ± 1.60
		Desi	107.8 ^{b} ± 15.8	89.6 ^{b} ± 7.8	22.9 ^{b} ± 14.3	196.6 ^{b} ± 33.7	107.0 ^{b} ± 33.7	73.8 ^{a} ± 1.83
Year								
		2005	128.1 ^{b} ± 24.8	93.4 ^{b} ± 14.1	34.2 ^{a} ± 15.3	243.9 ^{a} ± 46.5	150.6 ^{a} ± 35.4	72.8 ^{a} ± 1.60
		2006	135.0 ^{a} ± 32.6	110.2 ^{a} ± 19.7	29.4 ^{b} ± 13.3	230.6 ^{a} ± 50.5	120.5 ^{b} ± 33.6	72.4 ^{a} ± 1.83

Means (±SD) of triplicate analysis per year and all data on dry weight basis

Means followed by the same letter within a column and section do not differ significantly ($P < 0.05$)

[1] Biotype: K = Kabuli, D = Desi ; *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles

4.1.3.2.2 Thermal stability of chickpea starches

DSC was used to study the thermal properties of the chickpea starches compared with native potato starch (NPS, 91.0% total starch, 26.9% amylose)(Figure 4.1.16 and Table 4.1.24). The "biotype" effect of Kabuli and Desi starch was not significant for thermal properties. There was not 'year' effect for the enthalpy of starch gelatinization, but chickpea starches prepared from the 2006 harvest had much higher T_o (5.0°C higher) and T_p (4.1°C higher) values than did starches from the 2005 harvest (Figure 4.1.16). This was also observed earlier for DSC of the corresponding flours (starch gelatinization peak) (section 4.1.1.5.2). Since the amylose contents of starches from the 2005 and 2006 harvests were the same, these differences may be due to the influence of the molecular architecture of the crystalline region of corresponding starches (Noda, Takahata, Sato, Ikoma & Mochida, 1996; Singh et al., 2004).

The onset gelatinization temperatures ranged from 53.8 to 56.3°C for chickpea starches, whereas, it was 59.1°C for native potato starch. Compositional analysis of chickpea and NPS showed that chickpea starches had higher amylose content than that of the NPS. Sasaki, Yasui & Matsuki (2000) suggested that starches with higher amylose contents had more amorphous region and less crystalline, thereby lowering the gelatinization temperatures. However, peak temperature (T_p) and enthalpy of gelatinization (ΔH) of native potato starch were higher than those of the chickpea starches. Therefore, a lower temperature and lower energy are needed to break the intermolecular bonds in starch granules of chickpea starches to achieve gelatinization comparable to that of native potato starch.

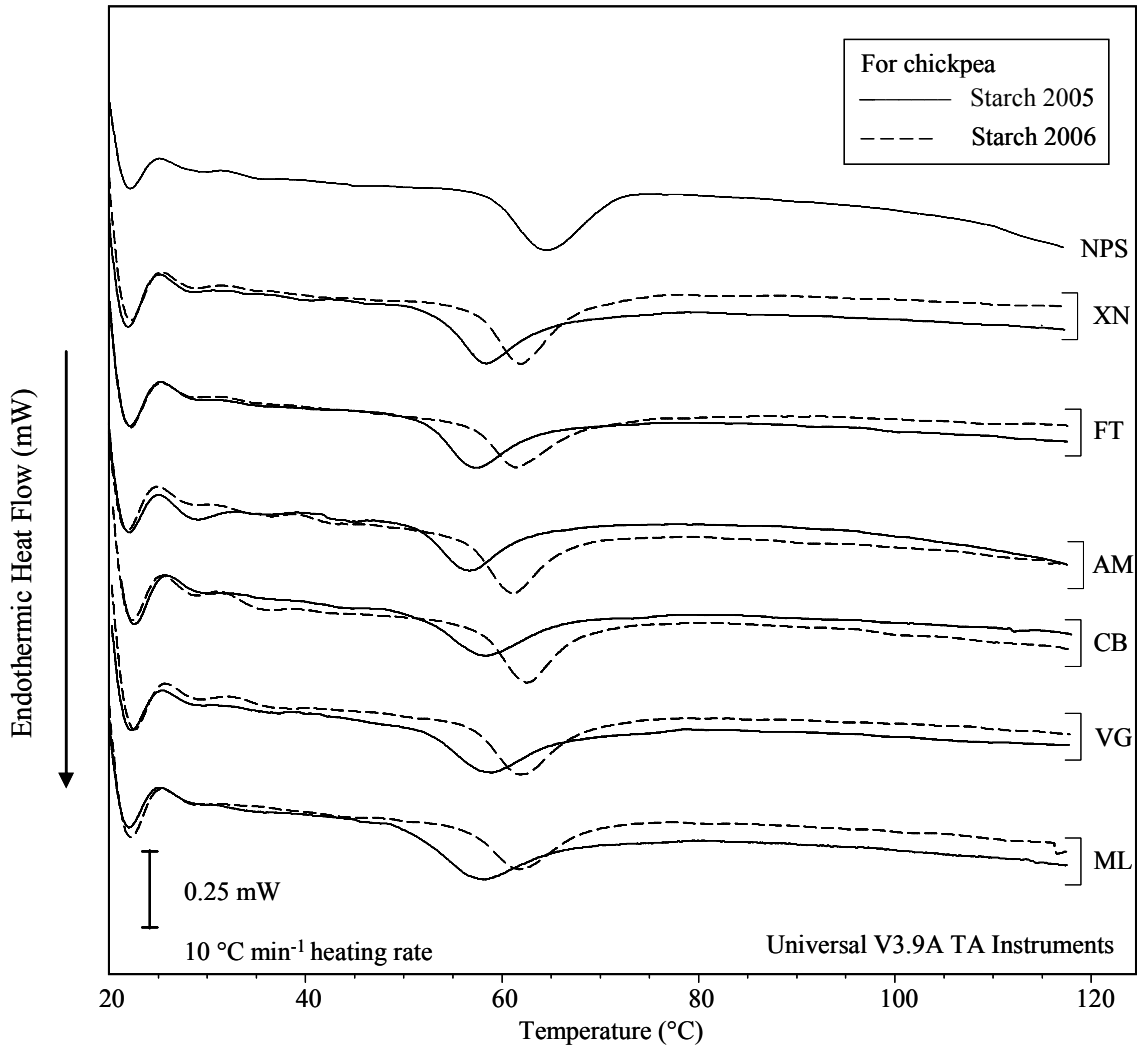

Figure 4.1.16 DSC thermal curves for various chickpea starches from 2005 and 2006 harvesting years. *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles; NPS = native potato starch

Table 4.1.24 Cultivar and year effects on thermal properties of starch from different chick-pea cultivars compared with native potato starch.

Var.	year	type[1]	T_o (°C)	T_p (°C)	ΔH(J/g)
Cultivar					
XN	05/06	K	56.29 [a] ± 2.10	60.60 [a] ± 1.88	2.70 [b] ± 0.08
FT	05/06	K	53.82 [a] ± 2.87	58.80 [a] ± 2.45	2.10 [cd] ± 0.15
AM	05/06	K	54.77 [a] ± 3.10	59.79 [a] ± 2.86	1.99 [d] ± 0.16
CB	05/06	D	54.99 [a] ± 3.09	60.32 [a] ± 2.23	2.43 [bc] ± 0.29
VG	05/06	D	54.28 [a] ± 2.39	60.46 [a] ± 1.84	3.08 [a] ± 0.22
ML	05/06	D	54.38 [a] ± 3.01	60.25 [a] ± 2.07	2.29 [cd] ± 0.25
NPS	-	-	59.09 ± 0.16	64.69 ± 0.31	3.87 ± 0.32
Biotype					
		Kabuli	54.96 [a] ± 1.25	59.67 [a] ± 1.16	2.27 [a] ± 0.38
		Desi	54.55 [a] ± 0.39	60.34 [a] ± 0.11	2.60 [a] ± 0.42
Year					
		2005	51.79 [b] ± 0.88	57.70 [b] ± 0.81	2.34 [a] ± 0.42
		2006	56.79 [a] ± 0.54	61.78 [a] ± 0.45	2.50 [a] ± 0.42

Means (±SD) of triplicate analysis per year
Means followed by same letter within a column and section do not differ significantly ($p < 0.025$)
[1] Biotypes: K = Kabuli, D = Desi. *Var:* XN = CDC Xena, FT = CDC Frontier, AM = Amit, CB = CDC Cabri, VG = CDC Vanguard, ML = Myles; NPS = native potato starch
T_o = onset temperature, T_p = peak temperature and ΔH = enthalpy change of gelatinization or denaturation

4.1.3.3 Conclusions

The proximate composition of native chickpea starches showed high purity (93-98%) and low residual protein (0.4-0.5%). The amylose contents (31.0-36.0%) and pasting temperatures (70.2-74.8°C) were comparable to those of other legume starches. Thermograms showed lower gelatinization temperatures for chickpea starches (58.8-60.6°C) than that of native potato starch (64.7°C), suggesting that chickpea starch may be a useful alternative to native potato starch in industrial thermally processed foods.

4.1.4 Overall summary of Study 1 and variety selection for meat applications

Six chickpea varieties were analyzed for their proximate, functional and thermal properties. Generally all chickpea flours were a rich source of protein and starch. Significant differences between the properties of seeds, flours, protein isolates and starches from Kabuli and Desi chickpea cultivars were observed. Seeds of Kabuli-type chickpea were significantly different from Desi-type chickpea seeds in their seed weight, seed coat, hydration capacity and swelling capacity. For flours, the two biotypes differed in their lightness value, protein, amylose, insoluble dietary fibre, and total dietary fibre contents, least gelation concentration, water holding capacity, oil absorption capacity, emulsion activity and stability indices. Protein isolates from Kabuli biotypes had significantly different crude fat, G″, onset temperature, WHC, OAC, EAI and ESI when compared to protein isolates from Desi-type chickpea. Starches from Kabuli-types were significantly different from Desi-type starches in their fat content and pasting properties. Therefore, one chickpea variety from each biotype (i.e. Kabuli and Desi) was selected for the second phase of study. Further screening within biotype was done according to their overall performance (seed, flour, isolates, and starch) in various chemical, thermal and functional properties. According to the findings, CDC Xena (Kabuli-type) and Myles (Desi-type) generally had higher numbers of desirable properties with respect to meat applications. Therefore, these two varieties were used for the second study, which involved evaluation of chickpea flour, starch and protein fractions in low-fat pork bologna.

4.2.1 Study 2: Chickpea flour as an extender in low-fat pork bologna

4.2.1.1 Proximate composition and CIE colour values of chickpea, pea & wheat flours

Flour additives were different in their chemical composition and colour (Table 4.2.1). Chickpea and pea flour had higher protein, fat and ash content than wheat flour. Chickpea flours had higher petroleum ether extractable lipids than the pea and wheat flour. Colour of the flour additives varied. Kabuli flour, having the highest L*, was lighter in colour than the other flours. Values for a* and b* of wheat flour indicated that it was greener and less yellow than chickpea and pea flour.

Table 4.2.1 Proximate composition[a] and colour values of flour additives

Flour binder	Mois-ture (%)	Protein (%)	Fat (%)	Ash (%)	CIE Colour[1]		
					L*	a*	b*
Kabuli	7.5 ± 0.6	22.0 ± 0.0	5.4 ± 0.0	2.7 ± 0.1	84.9±0.6	5.9±0.3	21.3±0.6
Desi	7.2 ± 0.1	23.0 ± 0.0	4.4 ± 0.0	3.1 ± 0.0	81.1±0.1	6.9±0.1	23.7±0.2
Wheat	7.8 ± 0.0	14.1 ± 0.1	0.4 ± 0.0	0.6 ± 0.2	81.0±0.8	2.5±0.1	7.9±0.2
Pea	9.5 ± 0.5	22.5 ± 0.1	1.1 ± 0.0	2.6 ± 0.0	81.9±0.4	6.7±0.0	23.9±0.1

[a] Mean (± SD) of duplicate determinations (reported on an as is basis)
[1] CIE colour: "L*" = lightness; "a*" = redness; "b*" = yellowness

4.2.1.2 Raw batter properties

Batter viscosity is a measure of the resistance of the meat emulsion to flow. The type and level of flour affected ($P<0.05$) viscosity of low-fat pork batter samples (Table 4.2.2). Addition of Kabuli, Desi and pea flour at 5.0% resulted in significantly higher ($P<0.05$) viscosity values than that of the control (no binder) and the 2.5% level of flour. The 2.5% level of addition resulted in similar or lower viscosity in the meat batter as the control. Flour addition at high levels may help to maintain the viscosity of the raw meat emulsion. According to Claus & Hunt (1991) and Claus, Hunt, Kastner, & Kropf (1990) more water and reduced-fat level can

lead to decreased batter viscosity of the comminuted meat products in the absence of any plant-based extender. However, addition of wheat (13.4% protein and 71.2% starch), normal barley (11.8% protein and 66.3% starch) and waxy barley flours (13.0% protein and 61.2% starch) increased batter viscosity by 31%, 34.9% and 37.3% respectively, compared to the control (Shand, 2000). The low batter viscosity may cause handing problems in pre-stuffing and stuffing processes and addition of plant binders including chickpea, pea and wheat flour can reduce this problem, by increasing raw batter viscosity.

Temperature of the meat batter at various points during processing was measured. Initial temperature of the meat was -1 to 0°C for every batch. There was a significant effect ($P < 0.05$) of binder addition on final batter temperature (right after stuffing). This may be due to three things, 1): temperature of the treatments (flour, seasoning, salt, etc.) which were at the room temperature and may impart a temperature rise to the batter, 2): addition of flour which increases the friction of the system also may affect the batter temperature rise, and 3) the control treatment had the highest amount of ice water in its' formulation.

Table 4.2.2 Effect of flour binders on apparent viscosity of raw low-fat batters and batter temperature just after stuffing

Treatment		Viscosity	Batter temperature
Binder	Level (%)	(cps) × 10^5	(°C)
Control	0	2.33 ^{bc} ± 0.08	8.9 ^{c} ± 0.9
Chickpea flour			
Kabuli	2.5	2.35 ^{bc} ± 0.10	11.2 ^{b} ± 0.7
	5.0	2.63 ^{a} ± 0.08	12.3 ^{ab} ± 0.9
Desi	2.5	2.03 ^{d} ± 0.12	11.9 ^{ab} ± 1.1
	5.0	2.66 ^{a} ± 0.08	12.9 ^{ab} ± 0.4
Wheat flour	2.5	2.00 ^{d} ± 0.15	12.7 ^{ab} ± 0.9
	5.0	2.46 ^{ab} ± 0.11	13.8 ^{a} ± 0.9
Pea flour	2.5	2.18 ^{cd} ± 0.09	12.7 ^{ab} ± 0.7
	5.0	2.69 ^{a} ± 0.09	13.3 ^{a} ± 0.2

$^{a-d}$ Means (± SD) within the same column with the same letter are not significantly different at $P < 0.05$
cps – centipoise unit

4.2.1.3 Chemical composition and pH of the cooked bologna

Mean values for proximate composition of the LFPB with added flours are shown in Table 4.2.3. Flour added LFPB had lower moisture content than the control which had no binder in the formulation. With increasing levels of flour from 2.5% to 5.0%, moisture content significantly decreased ($P < 0.05$) by approximately 77% to 75% respectively. This may be because water was replaced from the flour in the LFPB formulation. Protein contents of LFPB ranged from 13.9 to 14.7%, with a slight variation among the formulations. These would meet Canadian standards for protein content in cooked sausages; minimum of 9.5% meat protein, 11% total protein. Lecomte, Zayas & Kastner (1993) reported that addition of soy proteins in the meat batters increased the protein content of the product. However, Dzudie, Scher, & Hardy (2002) reported that with increasing levels of added common bean flour in beef sausage, protein content of the sausages decreased due to the composition difference.

Furthermore, Brown and Zayas (1990) concluded that the decrease in protein content of beef patties when the level of corn germ flour increased in the formulations was due to an increase in the carbohydrate content of the products. Fat content of flour added LFPB significantly ($P < 0.05$) differed from each other. Moreover, addition of flour to the LFPB only had minor effects on ash contents. No significant effects on pH were found among formulations. Similarly, Minrich et al. (1991) did not find any significant differences in pH of ground beef mixtures with wild rice flour.

Table 4.2.3 Proximate analysis of LFPB formulated with two levels and four types of flour binders

Treatment			Protein	Fat	Ash	
Binder	Level (%)	Moisture (%)	(%)	(%)	(%)	pH
Control	0	79.7 ^{a} ± 0.62	13.9 ^{bc} ± 0.40	2.6 ^{c} ± 0.92	2.7 ^{bc} ± 0.03	6.16 ^{a} ± 0.12
Chickpea flour						
Kabuli	2.5	77.1 ^{b} ± 0.51	14.2 ^{abc} ± 0.35	3.9 ^{ab} ± 0.95	2.8 ^{ab} ± 0.05	6.31 ^{a} ± 0.28
	5.0	75.4 ^{c} ± 0.57	14.3 ^{ab} ± 0.29	4.0 ^{ab} ± 0.75	2.9 ^{a} ± 0.03	6.31 ^{a} ± 0.13
Desi	2.5	77.0 ^{b} ± 0.61	14.1 ^{bc} ± 0.16	4.6 ^{a} ± 0.45	2.8 ^{a} ± 0.08	6.33 ^{a} ± 0.22
	5.0	75.8 ^{c} ± 0.82	14.4 ^{ab} ± 0.20	3.4 ^{bc} ± 1.32	2.9 ^{a} ± 0.05	6.32 ^{a} ± 0.07
Wheat flour	2.5	77.4 ^{b} ± 0.67	14.2 ^{abc} ± 0.30	3.4 ^{bc} ± 0.67	2.7 ^{c} ± 0.02	6.14 ^{a} ± 0.10
	5.0	75.2 ^{c} ± 0.22	13.8 ^{c} ± 0.94	3.4 ^{bc} ± 0.54	2.8 ^{ab} ± 0.16	6.18 ^{a} ± 0.12
Pea flour	2.5	77.1 ^{b} ± 1.24	14.1 ^{bc} ± 0.41	3.8 ^{ab} ± 1.56	2.7 ^{bc} ± 0.01	6.30 ^{a} ± 0.25
	5.0	75.4 ^{c} ± 0.32	14.7 ^{a} ± 0.28	3.5 ^{bc} ± 0.61	2.8 ^{ab} ± 0.03	6.23 ^{a} ± 0.13

$^{a-d}$ Means (± SD) within the same column with the same letter are not significantly different at $P < 0.05$

4.2.1.4 Cook yield, expressible moisture and purge losses

Table 4.2.4 shows effect of flour binders on cook yield and water holding parameters of cooked LFPB. Among the flour levels, 2.5% of Desi chickpea, pea and wheat flours resulted in similar cook yield as the control (Table 4.2.4). But the cook yield was significantly ($P < 0.05$) higher for low-fat bologna with 2.5% Kabuli flour or 5.0% levels of all the flours used than the control. The highest cook yield was observed for bologna with Kabuli, Desi and wheat flour at 5.0% level. Flours from the legumes and cereals contain mainly protein and starch (Table 4.2.3). Starch and proteins are biological macromolecules which can form gel matrices. Even in the presence of meat, they could form complex 3D gel network involving various forces such as van der Waals, electrostatic and hydrogen, which trapping fine particles of emulsified meat. This gel complex might help to retain water and fat during the cooking process.

Table 4.2.4 Effect of flour binders on cook yield, purge losses and expressible moisture of cooked low-fat pork bologna

Treatment		Cook yield	Expressible	Purge losses
Binder	Level (%)	(%)	moisture (%)	(%)
Control	0	94.95^{d} ± 0.49	17.33^{a} ± 0.78	3.76^{a} ± 0.34
Chickpea flour				
Kabuli	2.5	95.95^{bc} ± 0.34	12.70^{bc} ± 1.21	2.12^{b} ± 0.19
	5.0	97.20^{a} ± 0.50	10.28^{cd} ± 0.65	0.90^{cd} ± 0.23
Desi	2.5	94.73^{bcd} ± 0.56	13.49^{b} ± 1.28	2.20^{b} ± 0.20
	5.0	96.53^{ab} ± 0.47	11.19^{bcd} ± 0.98	1.33^{c} ± 0.19
Wheat flour	2.5	95.62^{bcd} ± 0.35	12.90^{bc} ± 0.97	1.36^{c} ± 0.23
	5.0	97.41^{a} ± 0.36	9.55^{d} ± 1.31	0.64^{d} ± 0.10
Pea flour	2.5	95.25^{cd} ± 0.05	13.83^{b} ± 1.05	1.92^{b} ± 0.34
	5.0	95.96^{bc} ± 0.60	12.52^{bc} ± 0.70	1.30^{c} ± 0.15

$^{a-d}$ Means (± SD) within the same column with the same letter are not significantly different ($P > 0.05$)

Chin et al. (1998) reported that low-fat bologna with konjac flour blends had higher cook yield than the control. Similar findings were reported for common bean flours in sausages

(Dzudie et al., 2002), wild rice flour in ground beef mixture (Minerich et al., 1991) and corn germ protein flour in broiled beef patties (Brown & Zayas, 1990). However, addition of 4% wheat and barley flour in ultra-low fat bologna (< 1% fat) did not affect the cook yield (Shand, 2000).

Ease of water loss of cooked bologna were determined by a centrifugation method (expressible moisture: EM) and gravitational drip method (purge loss). EM for the control was very high when compared to all other treatments ($P < 0.05$). Purge loss for cooked LFPB ranged from 0.64 to 2.20%. These values were significantly ($P < 0.05$) lower than that of control. Purge loss is the ratio of weight of the sample after 14-days storage period to the initial weight of the sample. Again addition of flour level at 5.0% to LFPB showed lower purge drip than with 2.5% flour addition. Our results for EM and purge loss were much lower than results of Shand (2000). Discrepancy in the results could be due to differences in the product formulations and the fat levels.

Pearson correlation coefficients were calculated in order to find out the strength and direction of a linear relationship among raw batter viscosity, temperature, cooking yield, EM, and purge loss (Table 4.2.5). An inverse relationship between viscosity and released water was found. Hence, it can be safely said that high viscous batters form a good gel structure that has high water retaining ability. There is a negative correlation between batter temperature and water holding parameters. Cook yield showed very strong opposite relationship with EM and purge loss ($P< 0.01$). And finally, the purge loss of cooked LFPB increased ($r = 0.83$) with the EM. Perhaps EM can be used as an indicator of purge for the LFPB.

Table 4.2.5 Correlation coefficients (r) of raw batter properties, cooking yield, EM and purge loss of cooked LFPB (combined data, n=27)

	Batter temperature	Cook yield	EM	Purge loss
Viscosity	0.06	0.42*	-0.44*	-0.29
Batter temperature	1	0.14	-0.48*	-0.56**
Cooking yield		1	-0.69***	-067**
EM			1	0.83***
Purge loss				1

*, **, *** = Significant at P<0.05, 0.01 and 0.001, respectively.

4.2.1.5 CIE colour

Data for colour of the control bologna and extended bologna slices are presented in Table 4.2.6. The L* and a* values were not greatly affected by adding flour or changing the level of flour addition. Bologna formulations with pea flour (2.5%), wheat flour (5.0%) and Kabuli flour (2.5%) were significantly (P<0.05) lighter in colour than the treatment with 5.0% Desi flour. The colour of the Desi seed coat may confer this small difference in L* value of the cooked LFPB. Control bologna and that with Kabuli flour at 2.5% had higher a* (redness) values than that of the bologna with 5.0% Desi flour (P<0.05). Addition of flour up to 5.0% to formulations significantly increased the CIE yellowness (b*), compared with the control. The highest b* value was observed for LFPB containing 5.0% Desi flour.

Shand (2000) found that wheat and barley flour binders had only small effects on colour of ultra low-fat bologna. Dzudie et al. (2002) reported that addition of common bean flour (CBF) at four different levels (2.5, 5.0 7.5 and 10.0%) in beef sausages had a significant effect on the colour of the product. Beef sausages containing 5.0, 7.5 and 10.0% CBF were significantly (P<0.05) lighter and sausages with CBF levels at 7.5, 10.0% showed significantly (P<0.05) lower degree of redness. As well, beef sausages with CBF levels at 2.5, 5.0, 7.5 and 10.0% were significantly (P<0.05) more yellow than the sample without added CBF (Dzudie et al., 2002). Furthermore, Prinyawiwatkul et al. (1997) reported that chicken nuggets extended

with fermented cowpea and peanut flour had higher L*, a* and b* values compared to the control.

Table 4.2.6 Effect of different flour binders on colour of cooked low-fat bologna

Treatment		CIE colour[1]		
Binder	Level (%)	L*	a*	b*
Control	0	68.23 [ab] ± 0.19	17.41 [a] ± 0.08	10.91 [d] ± 0.05
Chickpea flour				
Kabuli	2.5	70.04 [a] ± 0.22	17.34 [a] ± 0.12	13.17 [c] ± 0.05
	5.0	69.28 [ab] ± 0.11	16.85 [ab] ± 0.08	14.05 [b] ± 0.06
Desi	2.5	68.84 [ab] ± 0.22	16.61 [ab] ± 0.14	14.08 [b] ± 0.10
	5.0	67.52 [b] ± 0.31	16.14 [b] ± 0.18	15.57 [a] ± 0.14
Wheat flour	2.5	69.20 [ab] ± 0.35	17.21 [ab] ± 0.29	12.38 [c] ± 0.17
	5.0	69.64 [a] ± 0.17	17.14 [ab] ± 0.13	12.96 [c] ± 0.08
Pea flour	2.5	69.81 [a] ± 0.20	17.07 [ab] ± 0.16	12.89 [c] ± 0.14
	5.0	69.41 [ab] ± 0.46	17.06 [ab] ± 0.26	14.08 [b] ± 0.15

[a-d] Means within the same column with the same letter are not significantly different (P > 0.05)
[1] CIE colour: "L*" = lightness; "a*" = redness; "b*" = yellowness

To study the colour fading property of cooked and vacuumed-packed bologna slices, CIE colour parameters were measured after 0, 7, 14 and 21 days of storage (Figure 4.21.1). The same bologna from each treatment and control was used during storage for colour estimation to avoid variation. Storage did not significantly (P<0.05) affect the CIE colour values. Candogan & Kolsarici (2003) reported an increase in redness and no change in lightness of low-fat beef frankfurters formulated with carrageenan with pectin during storage. Naveena, Muthukumar, Sen, Babji, & Murthy (2006) reported that chicken patties formulated with finger millet flour at 0, 2.5, 5.0, 7.5% did not affect (P<0.05) the L*, a* values, whereas b* values generally decreased with increasing storage time (up to three week storage time) with a few exceptions. So, LFPB with chickpea flour at 2.5 or 5.0% levels has stable colour when it is vacuum packed and stored at refrigerated condition for up to 21 days.

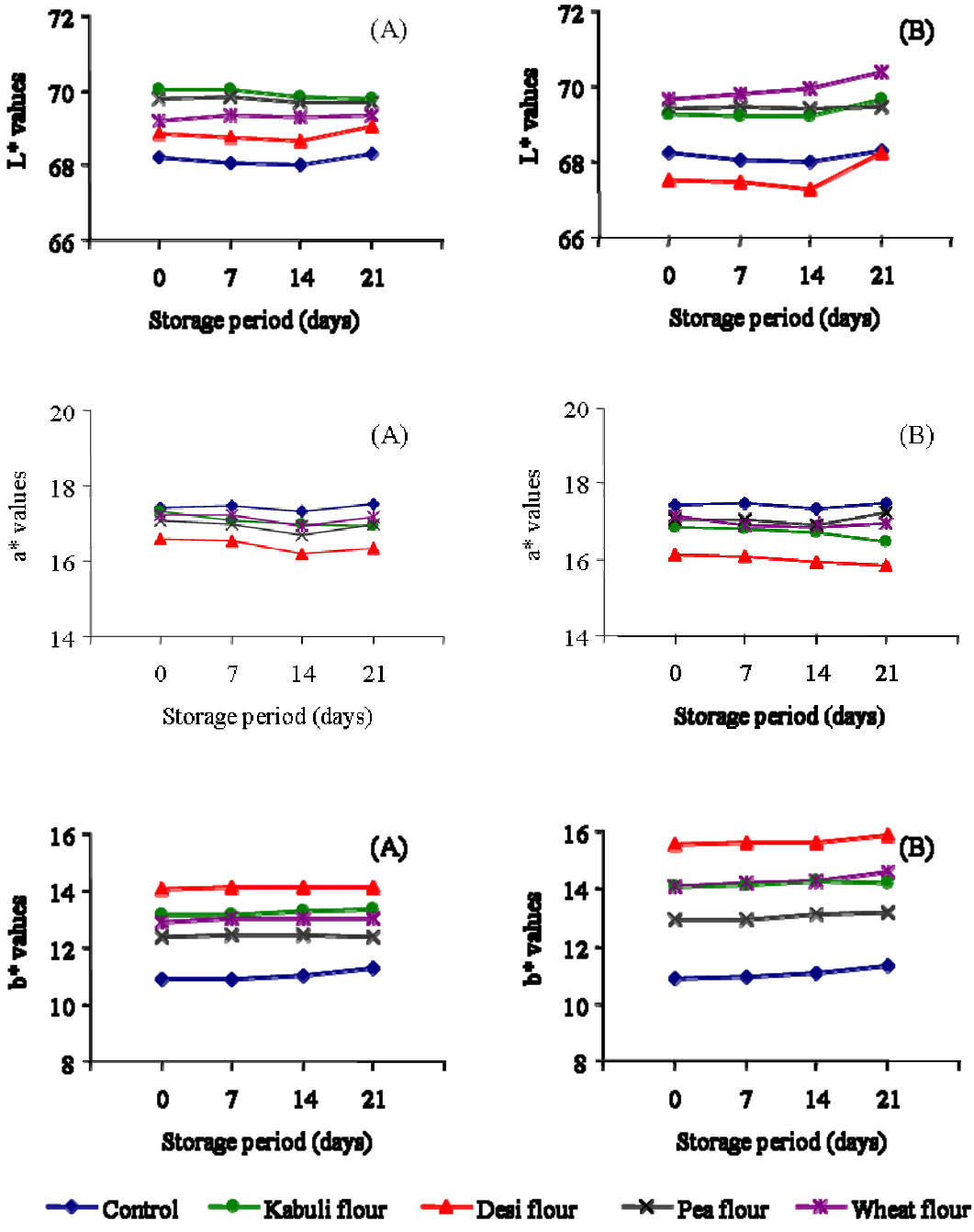

Figure 4.2.1 Changes in CIE colour values (L* [lightness], a* [redness] and b* [yellowness]) of cooked LFPB with 2.5 (A) or 5.0% (B) flour during storage at 4°C.

4.2.1.6 Allo-Kramer shear, texture profile analysis and torsion analysis.

Allo-Kramer (A-K) shear force values (N/g) of LFPB with different flour binders at 2.5 and 5.0% level are shown in Table 4.2.7. A-K Shear forces were similar ($P<0.05$) in cooked bologna with 2.5% Kabuli and Desi flour as compared with the control, while 2.5% wheat and pea flour containing LFPB were softer than the control. Bologna with Kabuli and Desi flour at 5.0% showed the highest ($P<0.05$) A-K shear forces among the treatments while bologna with 5.0% wheat and pea flour appeared to be intermediate in A-K shear force when compared to the control. A-K shear values increased as the level of chickpea flour in the formulation increased suggesting a strong gel structure formation in between meat and flour components. In contrast to effects seen with chickpea, a previous study showed that increased level of konjac flour gel had an inverse relationship with A-K shear force (Osburn & Keeton, 1994).

Means for instrumental texture profile analyses (TPA) are shown in Table 4.2.7. The control treatment was the least springy and chewy, while addition of 5.0% Desi flour and 5.0% pea flour produced bologna with the hardest texture (32% and 23%, higher than the control, respectively). Moreover, TPA-hardness value of formulations with 2.5% flour levels did not differ ($P<0.05$) from the control. However legume flour formulated LFPB (chickpea and pea) had significantly higher hardness and cohesiveness values than the control and wheat flour containing bologna at both addition levels. This might be due to the difference of protein content between the legume flours and wheat flour. Addition of any flour to the LFPB had no effect ($P<0.05$) on adhesiveness of the final product.

Shand (2000) compared the TPA parameters of ultra-low-fat pork bologna with added carrageenan (2.5%), soy protein concentrate (1%), potato starch (4%), wheat flour (4%) and barley flour (4%) and found that addition of carrageenan and soy protein concentrate had minor effects on texture, while use of wheat flour and barley flour significantly increased the

Table 4.2.7 Effect of different flour binders on textural properties of cooked low-fat bologna

Treatment		Allo-Kramer	Texture profile analysis				
Binder	Level (%)	Shear (N/g)	Hardness (N)	Adhesiveness (N)	Cohesiveness[1] -	Springiness (%)	Chewiness N mm
Control	0	14.4 ^{b} ± 0.7	103.7 ^{de} ± 5.2	-0.76 ^{a} ± 0.21	0.35 ^{cd} ± 0.04	73.1 ^{b} ± 2.4	455.5 ^{c} ± 8.5
Chickpea flour							
Kabuli	2.5	14.5 ^{b} ± 0.5	113.6 ^{cde} ± 8.3	-0.93 ^{a} ± 0.19	0.47 ^{a} ± 0.07	82.7 ^{a} ± 3.7	498.0 ^{bc} ± 9.0
	5.0	16.1 ^{a} ± 0.5	122.1 ^{bc} ± 8.3	-1.02 ^{a} ± 0.21	0.45 ^{ab} ± 0.05	81.0 ^{a} ± 1.3	587.0 ^{ab} ± 6.8
Desi	2.5	14.6 ^{b} ± 0.7	115.3 ^{cd} ± 6.4	-0.92 ^{a} ± 0.28	0.48 ^{a} ± 0.08	81.4 ^{a} ± 3.7	570.2 ^{ab} ± 9.3
	5.0	16.3 ^{a} ± 0.9	137.0 ^{a} ± 5.8	-0.97 ^{a} ± 0.18	0.48 ^{a} ± 0.07	79.4 ^{a} ± 5.5	624.4 ^{a} ± 8.6
Wheat flour	2.5	12.7 ^{c} ± 0.7	100.3 ^{e} ± 6.8	-0.77 ^{a} ± 0.22	0.30 ^{d} ± 0.05	79.9 ^{a} ± 2.9	359.6 ^{d} ± 7.5
	5.0	14.6 ^{b} ± 0.9	107.0 ^{de} ± 9.9	-1.03 ^{a} ± 0.21	0.31 ^{d} ± 0.06	80.5 ^{a} ± 2.8	364.0 ^{d} ± 3.1
Pea flour	2.5	13.0 ^{c} ± 0.8	108.1 ^{de} ± 6.8	-0.91 ^{a} ± 0.27	0.45 ^{ab} ± 0.04	80.7 ^{a} ± 3.3	553.8 ^{ab} ± 8.6
	5.0	15.4 ^{ab} ± 1.3	127.7 ^{ab} ± 4.4	-0.96 ^{a} ± 0.34	0.39 ^{bc} ± 0.05	80.6 ^{a} ± 2.8	505.0 ^{bc} ± 7.6

$^{a-e}$ Means within the same column with the same letter are not significantly different (P > 0.05)

[1] This value is dimensionless.

hardness of the bologna as compared to the control. Our results also differed from Dzudie et al. (2002) who reported that beef sausages with common bean flour (21.2% protein and 69.7% carbohydrates) addition at 2.5, 5.0, and 7.5% had lower shear force and hardness values.

Differences ($P < 0.05$) were found in shear stress and shear strain values of LFPB due to flour levels and type of flour added (Table 4.2.8). The highest shear stress (43.6 kPa) were observed for bologna with 5.0% Desi flour, whereas the lowest values were noted for the control (30.5 kPa) and 2.5% pea flour added LFPB (31.5 kPa). Except the pea flour containing bologna, 5.0% flour addition to bologna resulted in significantly ($P < 0.05$) higher shear stress values than that with 2.5% flour addition. Shear strain represents the elasticity of gelled meat products. Low fat bologna formulated with any flour had lower ($P < 0.05$) shear strain values than that the control. Differences ($P < 0.05$) were found in torsion rigidity values (shear stress/shear strain values) within the two levels of flour addition and different source of flours (Table 4.2.8 & Figure 4.2.2).

Table 4.2.8 Effect of different flour binders on torsion parameters of cooked low fat bologna

Treatment		Torsion shear values		
Binder	Binder level (%)	Stress (kPa)	Strain[1]	Rigidity (kPa)
Control	0	30.45 ^{f} ± 0.21	1.44 ^{a} ± 0.01	21.20 ^{f} ± 0.20
Chickpea flour				
Kabuli	2.5	32.53 ^{de} ± 0.19	1.07 ^{f} ± 0.01	30.30 ^{bc} ± 0.20
	5.0	37.02 ^{b} ± 1.14	1.25 ^{d} ± 0.02	29.55 ^{bcd} ± 1.28
Desi	2.5	34.81 ^{c} ± 0.77	1.28 ^{cd} ± 0.02	27.21 ^{ed} ± 0.96
	5.0	43.59 ^{a} ± 0.81	1.30 ^{bc} ± 0.01	33.45 ^{a} ± 0.50
Wheat flour	2.5	33.32 ^{cd} ± 1.12	1.08 ^{f} ± 0.05	30.94 ^{b} ± 1.18
	5.0	37.12 ^{b} ± 1.69	1.33 ^{b} ± 0.02	27.85 ^{d} ± 1.59
Pea flour	2.5	31.46 ^{ef} ± 0.28	1.25 ^{d} ± 0.01	25.10 ^{e} ± 0.28
	5.0	32.85 ^{de} ± 0.72	1.16 ^{e} ± 0.05	28.27 ^{cd} ± 1.42

[a-e] Means within the same column with the same letter are not significantly different ($P>0.05$)
[1] Shear strain is dimensionless.

The flour addition increased the torsion rigidity (increased shear stress and decreased shear strain) ($P<0.05$) when compared with the control.

The control treatment had a more "rubbery" texture than all other treatments (Figure 4.2.2). For all the flours tested the increased level of addition from 2.5 to 5.0% had an effect on texture. Texture of LFPB moved closer to "brittle" with wheat, pea and Desi flour whereas texture of Kabuli moved closer to "tough". The findings suggest that when flour addition was increased from 2.5 to 5.0% in the formulation, changes of torsion rigidity of wheat and Kabuli flour added bologna were driven by shear stress. Similarly pea flour added bologna showed torsion rigidity driven by the changes in shear strain while shear strain and shear stress were responsible for the changes of torsion rigidity in Desi flour added LFPB.

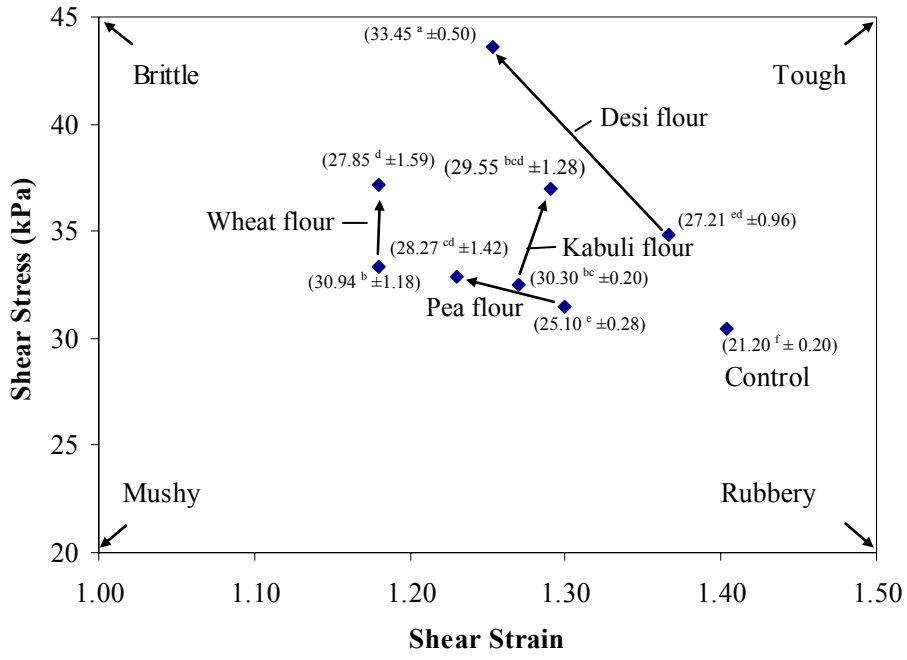

Figure 4.2.2 Torsion texture map of low-fat bologna containing different flours. Arrows point from the 2.5% flour to the corresponding 5.0% flour. Corresponding torsion rigidity values (shear stress/shear strain) (kPa) are given in parentheses. $^{a-f}$ Means of torsion rigidity with the same letter are not significantly different ($P>0.05$).

Table 4.2.9 shows the correlation coefficients among the textural parameters measured by TPA, Allo-Kramer and torsion geometry. Torsion stress had strong positive correlations with TPA hardness and A-K shear force values (r = 0.64). But, with torsion rigidity, only TPA hardness values were positively correlated (r = 0.51). Interestingly, torsion strain was negatively correlated with TPA springiness while torsion rigidity positively correlated with TPA springiness. There was a very strong correlation between A-K shear force and TPA-hardness (r = 0.71). Furthermore, TPA-hardness showed a strong positive correlation with TPA-cohesiveness and TPA-chewiness although TPA-adhesiveness had an opposite relationship (r = -0.41). The correlation of TPA-hardness and TPA springiness (r = - 0.58) was significant. Cohesiveness which measures the deformation of samples before it ruptures had a very strong positive (r = 0.83, $p < 0.001$) relationship with TPA-chewiness and a small positive (r = 0.42, $P < 0.05$) correlation with A-K shear values. Chewiness (the arithmetic result of multiplying the values of hardness, cohesiveness and springiness) was positively and significantly correlated with A-K shear force.

Table 4.2.9 Correlation coefficients (r) among torsion rigidity, TPA and A-K shear force values of cooked LFPB samples (combined data, n=27)

	2	3	4	5	6	7	8	9
1. Shear stress	0.14	0.71***	0.64***	-0.25	0.27	0.20	0.28	0.64***
2. Shear strain	1	-0.58**	-0.02	-0.00	-0.04	-0.50**	-0.04	0.25
3. Torsion rigidity		1	0.51**	-0.19	0.21	0.50**	0.23	0.34
4. Hardness			1	-0.41*	0.56**	0.28	0.55**	0.71***
5. Adhesiveness				1	-0.18	-0.58**	-0.13	-0.24
6. Cohesiveness					1	0.33	0.83***	0.42*
7. Springiness						1	0.18	0.08
8. Chewiness							1	0.44*
9. A-K shear								1

*, **, *** = Significant at p <0.05, 0.01 and 0.001, respectively.
1 – 3 Torsion shear test values; 4 – 8 TPA parameters; 9 – Allo-Kramer shear

4.2.1.7 Sensory properties

For determination of sensory properties of LFPB, 7 of the 9 treatments were evaluated. Only 5.0% wheat and pea flour added LFPB were used for comparison purposes. The main reason was to reduce the number of samples for panelists to limit panelist fatigue (seven samples at a time). Since commercial practice of flour in bologna is around 5.0% flour, we evaluated LFPB containing 5.0% wheat and 5.0% pea flour was evaluated as a legume flour comparison. Textural and flavour quality of cooked LFPB were evaluated by a 14-member semi-trained panel. Data are presented in Figure 4.2.3 and Table 4.2.10. The control had the highest initial juiciness while bologna with 5.0% wheat flour had the lowest value ($P < 0.05$). This may be due to the high water content in the control formulation (Meullenet, Chang, Carpenter, & Resurreccion, 1994; Shand, 2000) as flours were substituted for water (1:1) in other treatments. However, with continued chewing, there were no significant differences in sustained juiciness for Kabuli flour and 2.5% Desi flour formulations compared with the control, whereas, bologna with 5.0% Desi, wheat and pea flour had lower scores for sustained juiciness compared to the control. Saltiness was not significantly changed ($P > 0.05$) among each of the formulations.

At both flour levels, all the samples were perceived to be firmer ($P < 0.05$) than the control; 5.0% Desi treatment was scored as having the firmest texture. The results for TPA hardness showed that LFPB with 5.0% Kabuli and pea flour were the hardest product. However, panelists scored as the firmest products the bologna with 5.0% Kabuli and Desi flour and these scores were significantly higher than that of the 5.0% wheat or 5.0% pea flour added bologna. In the case of cohesiveness, bologna with 5.0% Desi flour had a higher value ($P < 0.05$) than the control and 2.5% Desi flour containing samples.

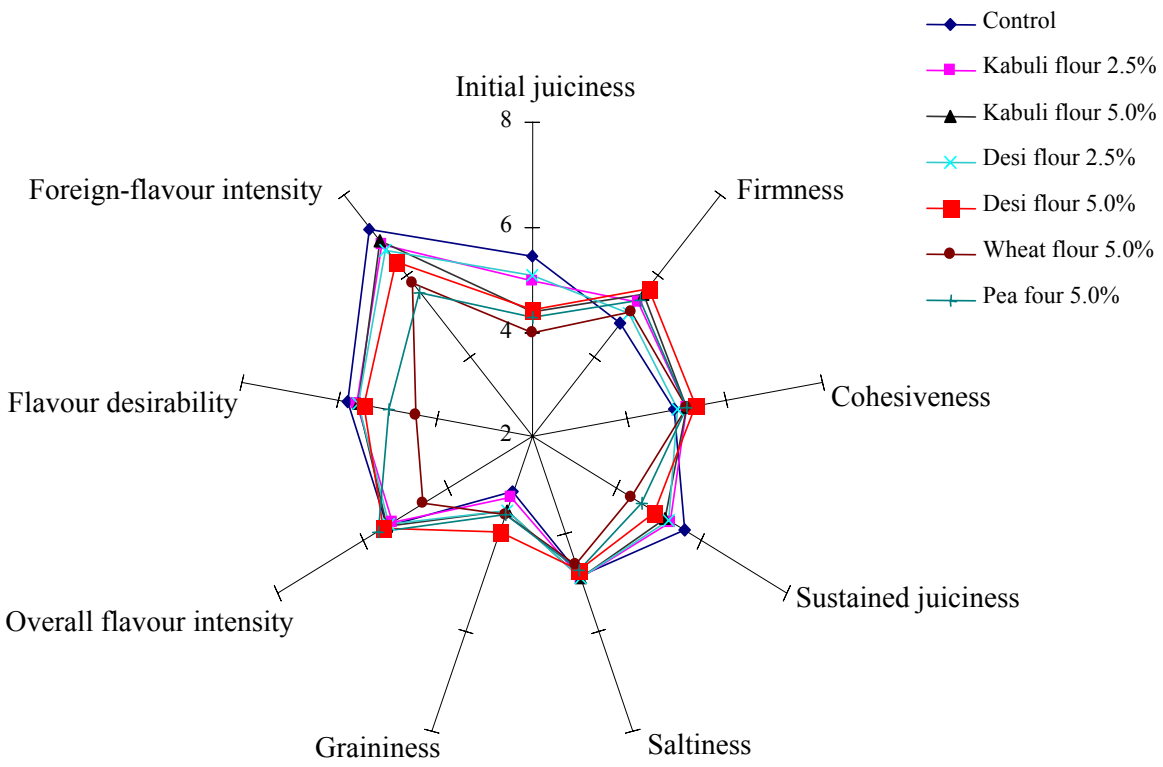

Figure 4.2.3 Sensory evaluation of LFPB formulated with Kabuli, Desi, pea and wheat flours at 2.5% and 5.0% levels. Initial and sustained juiciness: 8 (extremely moist) to 1 (extremely dry); firmness: 8 (extremely firm) to 1 (extremely soft); cohesiveness: 8 (extremely cohesive) to 1 (extremely brittle); saltiness: 6 (extremely salty) to 1 (not detectable); graininess 6 (extremely grainy) to 1 (not detectable); overall flavour intensity: 8 (extremely intense) to 0 (extremely band); flavour desirability: 8 (extremely desirable) to 1 (extremely undesirable); foreign flavour: 8 (no foreign flavour) to 1 (extremely intense foreign flavour)

Table 4.2.10 Effect of different flour binders on sensory properties of low-fat pork bologna[1]

Treatment		Initial	Firmness	Cohesiveness	Sustained	Graininess[2]
Binder	Level (%)	juiciness			juiciness	
Control	0	5.43 [a] ± 0.07	4.81 [d] ± 0.15	4.91 [b] ± 0.04	5.57 [a] ± 0.14	1.14 [c] ± 0.12
Chickpea flour						
Kabuli	2.5	4.97 [b] ± 0.05	5.36 [b] ± 0.07	5.17 [ab] ± 0.15	5.24 [ab] ± 0.11	1.24 [bc] ± 0.11
	5.0	4.39 [c] ± 0.06	5.50 [ab] ± 0.19	5.17 [ab] ± 0.04	5.11 [ab] ± 0.46	1.55 [ab] ± 0.27
Desi	2.5	5.08 [b] ± 0.08	5.07 [c] ± 0.12	5.01 [b] ± 0.08	5.23 [ab] ± 0.38	1.52 [ab] ± 0.18
	5.0	4.40 [c] ± 0.04	5.68 [a] ± 0.16	5.38 [a] ± 0.18	4.90 [bc] ± 0.08	1.93 [a] ± 0.14
Wheat flour	5.0	3.97 [d] ± 0.10	5.11 [c] ± 0.11	5.17 [ab] ± 0.08	4.31 [d] ± 0.42	1.60 [ab] ± 0.08
Pea flour	5.0	4.26 [c] ± 0.08	5.40 [b] ± 0.04	5.19 [ab] ± 0.23	4.55 [cd] ± 0.32	1.60 [ab] ± 0.36

[a-d] Means within the same column with the same letter are not significantly different (P>0.05)

[1] Highest possible score = 8 (extremely juicy, firm, cohesive, salty, intense, desirable, no foreign flavour); [1]lowest possible score = 1 (extremely dry, soft, brittle, bland, undesirable, intense) except for graininess or saltiness

[2] Highest possible score = 6 (extremely grainy or saltiness); [2] lowest possible score = 1 (no detectable graininess or saltiness)

Table 4.2.10 continued ….

Treatment			Saltiness[2]	Overall flavour intensity	Flavour Desirability	Foreign flavour intensity
Binder		Level (%)				
Control		0	2.86 [a] ± 0.07	5.40 [a] ± 0.18	5.86 [a] ± 0.26	7.14 [a] ± 0.14
Chickpea flour						
	Kabuli	2.5	2.88 [a] ± 0.08	5.31 [a] ± 0.15	5.64 [a] ± 0.31	6.81 [ab] ± 0.11
		5.0	2.90 [a] ± 0.22	5.43 [a] ± 0.12	5.59 [a] ± 0.27	6.88 [ab] ± 0.35
	Desi	2.5	2.88 [a] ± 0.18	5.38 [a] ± 0.08	5.60 [a] ± 0.18	6.64 [ab] ± 0.31
		5.0	2.74 [a] ± 0.08	5.52 [a] ± 0.18	5.48 [a] ± 0.15	6.36 [b] ± 0.25
Wheat flour		5.0	2.60 [a] ± 0.15	4.57 [b] ± 0.43	4.40 [c] ± 0.15	5.86 [c] ± 0.26
Pea flour		5.0	2.76 [a] ± 0.11	5.62 [a] ± 0.11	4.95 [b] ± 0.34	5.57 [c] ± 0.07

[a-d] Means within the same column with the same letter are not significantly different (P>0.05)
[1] Highest possible score = 8 (extremely juicy, firm, cohesive, salty, intense, desirable, no foreign flavour); [1]lowest possible score = 1 (extremely dry, soft, brittle, bland, undesirable, intense) except for graininess or saltiness
[2] Highest possible score = 6 (extremely grainy or saltiness); [2] lowest possible score = 1 (no detectable graininess or saltiness)

The lowest level of graininess ($P < 0.05$) was observed for the control and 2.5% Kabuli flour added bologna. The higher score for graininess in Desi flour containing LFPB may be due to the contained finely ground seed coat in the flour. Scores for flavour attributes such as overall flavour intensity and flavour desirability were very similar for all treatments, except 5.0% wheat flour and 5.0% pea flour (only in flavour desirability) formulated bolognas, which were lower ($P < 0.05$) for both traits. Foreign flavour intensity is the amount of any atypical or off-flavours present in the mouth after complete mastication, which was evaluated in order to determine if the beany flavour of the flour ingredients was perceived. Both chickpea formulations and the control bologna had very weak foreign flavour and did not significantly ($P > 0.05$) differ from each other. But panelists were able to identify ($P < 0.05$) a foreign flavour in 5.0% wheat and pea flour formulations giving a score of slightly weak to moderately weak. Our sensory data clearly shows that addition of Kabuli and Desi chickpea up to 5.0% in low-fat bologna was not detected by the panelists in terms of flavour.

Onweluzo et al. (2003) reported that addition of 0.5% *Detarium microcarpum* (Dm) flour in Buffalo meat loaves had a comparable taste score with the control while with 1.0%, Dm products had significantly ($P < 0.05$) lower scores for taste and overall acceptability suggesting that they were less preferred. Mean scores for overall rating of the ground beef mixtures containing 15 and 30% wild rice were not significantly different ($P > 0.05$) from the control (Minerich et al., 1991).

Modi et al. (2003) studied and compared the sensory quality of legume flours (soya bean, Bengal gram, green gram and black gram) (8%) in buffalo meat burgers. However, the burger with black gram dhal flour had better sensory quality attributes compared to other legumes. Moreover, Osburn & Keeton (1994) reported that 0, 10 and 20% konjac flour in low-fat prerigor fresh pork sausages did not significantly affect sensory properties such as livery, metallic, astringent, salt, sweet, and sour.

Table 4.2.11 shows the Pearson correlation analysis of the instrumental texture data and the sensory data. Generally, there were no important relationships between instrumental texture values and panelist perceptions, with a few exceptions. However, there was a strong positive correlation between torsion rigidity and sensory graininess (r = 0.73), TPA-hardness and graininess (r = 0.60), TPA-cohesiveness and sensory flavour desirability (r = - 0.58) and similarly TPA-chewiness with overall flavour intensity (r = - 0.65) and flavour desirability (r = - 0.58). However there were more correlations among sensory attributes than in between instrumental texture data and sensory data which were surprising. There was a very strong positive relationship of initial juiciness with sustained juiciness (r = 0.94) and sustained juiciness played a powerful role on perception of flavour desirability (r = 0.85). Interestingly, flavour desirability had a very strong and positive correlation with low foreign flavour intensity.

Table 4.2.11 Correlation coefficients (*r*) of the instrumental texture studies and sensory data (combined data, n=21)

	8	9	10	11	12	13	14	15	16
1. Torsion rigidity	-0.52*	0.32	0.33	-0.31	-0.30	0.73**	-0.05	-0.18	-0.21
2. TPA-Hardness	-0.31	0.18	0.42	-0.17	-0.14	0.60**	0.48*	0.05	-0.32
3. TPA-Adhesiveness	0.24	-0.45*	0.07	0.19	0.33	0.04	0.24	0.06	0.13
4. TPA-Cohesiveness	0.11	0.07	0.04	0.36	0.33	0.16	0.50*	0.57**	0.33
5. TPA-Springiness	-0.34	0.41	-0.03	-0.24	-0.13	-0.14	-0.24	-0.23	-0.23
6. TPA-Chewiness	0.11	0.05	0.17	0.36	0.41	0.08	0.65**	0.58**	0.24
7. A-K shear	-0.31	0.18	0.34	-0.13	0.08	0.47*	0.37	0.06	-0.07
8. Initial juiciness	1	0.27	0.03	0.94***	0.55*	-0.68**	0.33	0.72**	0.58**
9. Firmness		1	0.22	-0.19	-0.54*	0.18	-0.33	-0.25	-0.06
10. Cohesiveness			1	0.08	-0.26	0.17	0.13	-0.01	0.05
11. Sustained juiciness				1	0.56**	-0.59**	0.42	0.85***	0.72**
12. Saltiness					1	-0.32	0.56**	0.62**	0.39
13. Graininess						1	0.04	-0.44*	-0.45*
14. Overall flavour intensity							1	0.61**	0.17
15. Flavour desirability								1	0.76***
16. Foreign flavour intensity									1

*, **, *** = Significant at $p < 0.05$, 0.01 and 0.001, respectively.

4.2.1.8 Summary and conclusions

This study demonstrated that incorporation of chickpea flour at both levels (2.5 and 5.0%) into low-fat pork bologna increased the product's cook yield and decreased expressible moisture and purge significantly ($P < 0.05$). Compared to the control and bologna with 2.5% chickpea flour, adding 5.0% chickpea flour increased ($P < 0.05$) torsion shear stress, TPA (texture profile analysis) hardness, springiness, chewiness and Allo-Kramer shear values. Moreover Desi chickpea flour at 5.0% (DCF 5.0%) showed the highest TPA hardness, chewiness, Allo-Kramer and torsion shear stress. Furthermore bologna extended with 5.0% DCF had the highest b* and the lowest L* and a* values.

In general, addition of chickpea flours increased the sensory firmness, cohesiveness and graininess scores while the sensory scores for initial juiciness of the LFPB were decreased. For most flavour properties, bologna with Kabuli and Desi chickpea flour performed similarly to the control. However panellists noted more foreign-flavours with addition of wheat and pea flour at 5.0%. Bologna with chickpea flour were juicier and firmer than samples with added wheat flour. Results from this study indicated that chickpea flour at 2.5 and 5.0% has good potential as an extender in low-fat emulsion type meat systems.

4.2.2 Chickpea protein isolates as an extender in low-fat pork bologna

4.2.2.1 Protein isolate properties

The proximate composition of nonmeat proteins studied varied considerably, particularly with regard to protein and fat contents (Table 4.2.12). Both soy and pea protein isolates had higher protein content and lower petroleum ether extractable fat content than that of the chickpea protein isolates. These chickpea isolates, which were prepared by an ultrafiltration method had quite different chemical composition when compared to corresponding isolates prepared in the lab by using isoelectric precipitation (see Table 4.1.13). Also chickpea samples were from a more recent harvest year (2007).

Table 4.2.12 Proximate composition[a] of protein isolates

Protein Isolates	Moisture (%)	Protein (%)	Crude fat (%)	Ash (%)
Kabuli	6.3 ± 0.32	74.5 ± 0.2	9.8 ± 0.15	2.5 ± 0.19
Desi	3.7 ± 0.06	70.1 ± 0.7	6.2 ± 0.44	2.8 ± 0.17
Soy	4.3 ± 0.11	86.3 ± 0.0	0.5 ± 0.02	4.2 ± 0.07
Pea	5.3 ± 0.09	77.7 ± 0.0	2.8 ± 0.01	7.4 ± 0.86

[a] Mean (± SD) of duplicate determinations (reported on an as is basis)

4.2.2.2 Raw batter properties

Chickpea protein isolates (CPI) in low-fat pork bologna (LFPB) were characterized and compared with other extenders: soy protein isolate (SPI) and pea protein isolate (PPI). Protein levels and moisture contents of each LFPB formulation are shown in Table 4.2.13. LFPB were formulated with mainly three levels of total proteins; control-I had 11.1%, control-II and 3.0% plant protein had a total of 14.1% and finally 1.5% plant protein containing LFPB had total of 12.6% protein levels. All treatments had the same amount of meat protein (11.1%), except control-II which had 14.1%. Control-II had the lowest level of water among the formulations (17.9%) which was 48.6% lower than amount of water added to the control-I formulation.

Table 4.2.13 Protein levels in the each LFPB formulation manufactured with four types and two levels of protein isolates.

Binder[1]		Plant Protein (%)	Binder level (%)	Meat protein (%)	Ice/water (%)	Total protein (%)
Control	I	0	-	11.1	34.8	11.1
	II	0	-	14.1	17.9	14.1
CPI	Kabuli	1.5	1.97	11.1	32.8	12.6
		3.0	3.95	11.1	30.9	14.1
	Desi	1.5	2.03	11.1	32.7	12.6
		3.0	4.12	11.1	30.7	14.1
SPI		1.5	1.66	11.1	33.1	14.1
		3.0	3.33	11.1	31.5	12.6
PPI		1.5	1.83	11.1	33.0	12.6
		3.0	3.66	11.1	31.1	14.1

[1] CPI-chickpea protein isolate, SPI-soy protein isolate, PPI- pea protein isolate

The temperature of all ground meat samples were between -0.1 and 0°C at the beginning of processing of LFPB with the plant proteins. Temperature of each processing step was also monitored. Before the vacuum was applied, the average temperature of meat batters ranged from 7.8 to 11.4°C, after the vacuum procedure it ranged from 9.5 to 12.3°C. Batter temperature right after the final step (stuffing) of the LFPB processing were significantly affected among formulations (Table 4.2.14). Addition of plant proteins significantly ($P < 0.05$) increased the temperature of the LFPB. Despite the fact that control-II had the lowest level of ice water, its raw batter temperature was not different ($P > 0.05$) from the control-I.

Table 4.2.14 Effect of protein isolate extenders on apparent viscosity of raw low-fat batters and batter temperature right after stuffing

Treatment			Viscosity	Batter temperature
Binder[1]		Protein level (%)	cps × 10^{-5}	°C
Control	I	0	2.33 ^{cd} ± 0.04	10.5 ^{e} ± 0.1
	II	0	3.59 ^{a} ± 0.31	10.1 ^{e} ± 0.4
CPI	Kabuli	1.5	2.17 ^{cd} ± 0.09	11.5 ^{d} ± 0.1
		3.0	2.46 ^{c} ± 0.19	11.9 ^{cd} ± 0.0
	Desi	1.5	2.38 ^{cd} ± 0.25	11.9 ^{cd} ± 0.1
		3.0	2.47 ^{c} ± 0.09	12.6 ^{ab} ± 0.5
SPI		1.5	2.56 ^{c} ± 0.23	12.1 ^{bc} ± 0.5
		3.0	3.05 ^{b} ± 0.12	12.6 ^{a} ± 1.0
PPI		1.5	2.00 ^{d} ± 0.23	12.1 ^{abc} ± 0.1
		3.0	2.57 ^{c} ± 0.53	12.6 ^{ab} ± 0.1

$^{a-d}$ Means within the same column with the same letter are not significantly different (P > 0.05)
cps – centipoise unit
[1] Non meat protein : CPI-chickpea protein isolate, SPI-soy protein isolate, PPI- pea protein isolate

Some differences in batter viscosity were observed with addition of protein isolates (Table 4.2.14). Viscosity of control-II, containing the least amount of water and the most meat, was the highest. The second highest batter viscosity was noted for the sample containing 3.0% SPI while others were not significantly different (P > 0.05) from the viscosity of control-I. The control-II and 3.0% soy protein containing samples increased batter viscosity by 54 and 31%, respectively as compared to the control-I. Generally, batter viscosity was increased as protein level increased from 1.5 to 3.0%. Our results were in accordance with the findings of Shand (2000) who found that ultra-low fat bologna with 3.0% soy protein concentrate had significantly (P < 0.05) higher batter viscosity than the control (no binder). Gnanasambandam & Zayas (1992) reported that the batter viscosity of comminuted meat products with plant proteins (wheat germ, corn germ and soy protein) was significantly lower than that of the control 1 and significantly higher than the control 2 (control 1: 28.0% water, control 2: 31.5% water, and wa-

ter content of control 2 same as other treatments). Further, they found no difference in viscosity among treatment samples, whereas control 1 had higher viscosity than the control 2 ($P<0.05$).

4.2.2.3 Chemical composition and pH of the cooked bologna

Chemical composition of LFPBs is shown in Table 4.2.15. As expected, control-II had the lowest moisture and the highest protein content. Except for 3.0% PPI formulation, replacement of water with legume proteins had minimum effect on moisture content of the cooked product. LFPB with SPI at 3.0% had significantly higher pH values than that of the controls. The pH of the other formulations was around 6.2-6.3. Similar pH results for ultra low-fat (<1%) pork bologna with soy protein concetrate (1.0%) has been reported (Shand, 2000). Further, Shand (2000) reported that addition of soy protein isolates at 1.0% in ultra low-fat bologna increased the protein content and decreased the moisture content of cooked product when compared to the control (no binder).

Table 4.2.15 Proximate analysis of LFPB formulated with two levels and four types of protein isolate extenders

Treatment			Moisture	Protein	
Binder[1]		Protein level (%)	(%)	(%)	pH
Control	I	0	78.5 ^{a} ± 0.34	14.0 ^{c} ± 0.64	6.15 ^{b} ± 0.06
	II	0	75.0 ^{e} ± 0.77	16.5 ^{a} ± 0.52	6.18 ^{b} ± 0.12
CPI	Kabuli	1.5	78.0 ^{ab} ± 0.50	14.5 ^{bc} ± 1.39	6.23 ^{ab} ± 0.08
		3.0	76.4 ^{dc} ± 0.64	14.9 ^{bc} ± 0.86	6.24 ^{ab} ± 0.18
	Desi	1.5	77.7 ^{abc} ± 1.23	14.7 ^{bc} ± 0.60	6.30 ^{ab} ± 0.15
		3.0	77.1 ^{bcd} ± 0.97	15.0 ^{bc} ± 1.26	6.30 ^{ab} ± 0.14
SPI		1.5	78.2 ^{ab} ± 0.79	14.5 ^{bc} ± 0.43	6.23 ^{ab} ± 0.07
		3.0	77.5 ^{abc} ± 0.52	15.7 ^{ab} ± 0.24	6.34 ^{a} ± 0.09
PPI		1.5	77.8 ^{ab} ± 0.33	14.1 ^{c} ± 0.45	6.21 ^{ab} ± 0.17
		3.0	76.0 ^{ed} ±0.59	15.7 ^{ab} ± 0.53	6.21 ^{ab} ±0.09

$^{a-d}$ Means (± SD) within the same column with the same letter are not significantly different at $P < 0.05$

[1] Non meat protein : CPI-chickpea protein isolate, SPI-soy protein isolate, PPI- pea protein isolate

4.2.2.4 Expressible moisture, purge losses and cook yield

The cook yield reflects retention of water of the meat matrix during the cooking process (Table 4.2.16). Control-I (78.5% moisture content) had lower ($P < 0.05$) cooking yield than all other formulations except bologna with 1.5% Kabuli CPI and 3.0% PPI. Cook yield of the control-II were similar to LFPB having 3.0% Kabuli CPI and Desi CPI at both levels whereas it was significantly ($P < 0.05$) higher than the low fat bologna with 1.5 and 3.0% of SPI and PPI. Lecomte et al. (1993) reported that the comminuted meat containing soy protein had the highest cook yield (89.62%). Gnanasambandam & Zayas (1992) found the highest yield of frankfurters containing wheat germ protein whereas the lowest was for the control 2 (31.5% water).

Table 4.2.16 Effect of protein isolates on cook yield, purge losses and expressible moisture of cooked bologna

Treatment			Cook yield	Expressible moisture	Purge losses
Binder[1]		Protein level (%)	(%)	(%)	(%)
Control	I	0	$94.24^{f} \pm 0.79$	$17.64^{a} \pm 0.57$	$4.34^{a} \pm 0.26$
	II	0	$96.93^{a} \pm 0.48$	$9.40^{f} \pm 0.39$	$0.68^{h} \pm 0.12$
CPI	Kabuli	1.5	$94.58^{ef} \pm 0.55$	$14.48^{b} \pm 0.97$	$3.25^{c} \pm 2.15$
		3.0	$96.50^{ab} \pm 0.86$	$11.55^{e} \pm 0.94$	$1.65^{g} \pm 0.22$
	Desi	1.5	$96.05^{abc} \pm 0.18$	$13.11^{cd} \pm 0.98$	$2.47^{d} \pm 0.22$
		3.0	$96.65^{ab} \pm 0.29$	$10.17^{f} \pm 0.71$	$1.97^{f} \pm 0.32$
SPI		1.5	$95.79^{bcd} \pm 0.46$	$14.34^{bc} \pm 0.81$	$3.21^{c} \pm 0.23$
		3.0	$95.24^{cde} \pm 0.00$	$13.37^{bcd} \pm 0.98$	$2.16^{e} \pm 0.29$
PPI		1.5	$95.05^{cb} \pm 0.57$	$12.88^{d} \pm 0.89$	$3.55^{b} \pm 0.35$
		3.0	$95.05^{edf} \pm 0.21$	$12.61^{de} \pm 0.87$	$2.53^{d} \pm 0.35$

[a-d] Means within the same column with the same letter are not significantly different ($P > 0.05$)
[1] Non meat protein : CPI-chickpea protein isolate, SPI-soy protein isolate, PPI- pea protein isolate

Expressible moisture (EM) and purge loss of LFPB were significantly affected by incorporated protein additives (Table 4.2.16). Control-I, with highest water content and no bind-

ers in its formulation, had the highest EM value (17.6%). The lowest values for EM was observed for control-II (17.9% added water in the formulation) and bologna with 3.0% Desi CPI (30.7% water in the formulation) indicating Desi protein also can play a similar role of retention of water as did the extra meat in control-II. Treatments with 3.0% CPI protein showed a decrease in EM ($P < 0.05$) when compared with 1.5% CPI. Nevertheless, no differences ($P > 0.05$) in EM were found among treatments containing either level of added SPI or PPI; however 1.5% PPI had lower EM values than than of the 1.5% SPI.

Purge losses were significantly affected by addition of protein additives (Table 4.2.16). Purge losses of the control-II was the lowest. However, addition of plant proteins to LFPB had resulted in lower ($P < 0.05$) purge losses than the control-I. LFPB samples with 3.0% plant protein had slightly lower purge losses than the 1.5% level. Desi CPI inclusion at level 1.5% had better purge control than the SPI and PPI counterpart. Moreover, bologna with 3.0% Kabuli and Desi CPI had less purge than the LFPB with SPI and PPI. Similar results were reported by Lecomte et al. (1993), Gnanasambandam & Zayas (1992) and Kassama et al. (2003) where soya protein (1%), wheat germ protein, corn germ protein, and soy protein (3.5%) and soy protein (0, 2, 3.5 and 5%), respectively, in comminuted meat products increased the water holding capacity. However, opposite findings were reported by Shand (2000). She observed EM of ultra low-fat bologna (>1% fat) with 1.0% soy protein concentrate did not significantly ($P > 0.05$) differ from the control. She also noted that 1.0% soy protein concentrate made only a minor contribution to purge control. Similar findings to Shand (2000) for purge losses were reported by Chin et al. (1999) who formulated low-fat beef bologna with SPI (2 and 4% crude protein) and did not show any significant differences between the low-fat control and SPI treatments. Therefore, incorporation of chickpea protein at 1.5% into the low-fat bologna increased the water holding properties and cook yield when compared to the control-I and addition of 3.0% Kabuli and Desi protein at both levels are as effective as meat when compared to the control-II with regard to cook yield.

Viscosity of raw batter showed negative correlation with EM (r = -0.50, P < 0.01) and purge loss (r = -0.67, P < 0.001) of cooked LFPB (Table 4.2.17). Similar correlation between water binding properties and viscous properties of meat batters was reported by Gnanasambandam & Zayas (1992). As expected, EM and purge control were strongly and negatively correlated with cooking yield. Further, EM was an excellent indicator of purge loss (r = 0.86, P < 0.001).

Table 4.2.17 Correlation coefficients (r) of raw batter properties, cooking yield, EM and purge loss of cooked LFPB (combined data, n=27)

	Batter temperature	Cooking yield	EM	Purge loss
Viscosity	-0.33	0.35	-0.50**	-0.67***
Batter temperature	1	0.03	-0.13	-0.03
Cooking yield		1	-0.77***	-0.71***
EM			1	0.86***
Purge loss				1

*, **, *** = Significant at p <0.05, 0.01 and 0.001, respectively.

4.2.2.5 CIE colour

The addition of legume proteins to LFPB did not affect (P > 0.05) lightness (L*) and redness (a*) as compared to the control-I (Table 4.2.18). However, redness of 3.0% Desi formulations was significantly lower than the control-I. Control-II had lower L* value and higher a* (P < 0.05) than that of the other formulations, indicating higher redness and darker colour than other samples. This might be due to the high level of meat (80%)(more myoglobin) and low level of added water (18%)(less dilution) in its formulation.

Table 4.2.18 Effect of different protein isolates on colour of cooked bologna

Treatment			CIE colour		
Binder[1]		Level (%)	L*	a*	b*
Control	I	0	68.72 ^{ab} ± 0.22	17.63 ^{b} ± 0.21	11.18 ^{f} ± 0.12
	II	0	67.83 ^{b} ± 0.17	19.09 ^{a} ± 0.11	12.62 ^{d} ± 0.07
CPI	Kabuli	1.5	69.65 ^{a} ± 0.25	17.04 ^{bc} ± 0.12	13.36 ^{c} ± 0.12
		3.0	69.37 ^{ab} ± 0.25	16.92 ^{bc} ± 0.11	14.79 ^{b} ± 0.12
	Desi	1.5	68.41 ^{ab} ± 0.14	17.06 ^{bc} ± 0.08	14.63 ^{b} ± 0.08
		3.0	68.61 ^{ab} ± 0.05	16.42 ^{c} ± 0.10	17.23 ^{a} ± 0.08
SPI		1.5	68.71 ^{ab} ± 0.44	17.03 ^{bc} ± 0.24	12.13 ^{e} ± 0.15
		3.0	68.25 ^{ab} ± 0.39	17.35 ^{bc} ± 0.15	13.08 ^{c} ± 0.07
PPI		1.5	68.92 ^{ab} ± 0.24	17.35 ^{bc} ± 0.23	13.14 ^{c} ± 0.12
		3.0	69.54 ^{ab} ± 0.06	17.40 ^{b} ± 0.05	14.83 ^{b} ± 0.05

$^{a-d}$ Means within the same column with the same letter are not significantly different (P > 0.05)
[1] Non meat protein : CPI-chickpea protein isolate, SPI-soy protein isolates, PPI- pea protein isolates
[2] CIE colour: "L*" = lightness; "a*" = redness; "b" = yellowness

Formulations containing plant protein additives showed significantly (P < 0.05) higher b* values than the controls. 3.0% Desi CPI added samples had the highest yellowness scores among all other treatments. However, the addition of 1.5% Kabuli CPI yielded equivalent yellowness values to bologna with SPI and likewise bologna with 3.0% Kabuli and 1.5% Desi CPI had same yellowness values as that with 3.0% PPI. These results are in agreement with those of previous studies which showed control having low water content (control 1) to be darker and more red than the regular control 2 (high water containing control) and treatments with wheat germ protein, corn germ protein and soy protein were not different from the control 2 (Gnanasambandam & Zayas, 1992). However, in the present study, incorporation of legume protein isolates into low-fat bologna could increase the yellowness which may be undesirable from a consumer acceptance point of view.

4.2.2.6 Allo-Kramer shear, texture profile analysis and torsion analysis.

Table 4.2.19 shows data for texture of LFPB with added protein extenders. Significant differences ($P < 0.05$) were found in hardness, cohesiveness, chewiness and Allo-Kramer (A-K) shear force values within the two levels of added protein as well as compared with controls. Control-II had the highest ($P < 0.05$) hardness, adhesiveness, chewiness and A-K shear force values. These values were approximately double the values that obtained for the control-I. This clearly indicates that addition of more meat and having less water (3.5% less) had played a vital role in the structure development of the LFPB.

TPA-hardness scores for bologna with 1.5% Kabuli CPI, SPI, and PPI were same as the control-I whereas bologna with 3.0% Kabuli/Desi CPI, SPI, PPI and 1.5% Desi CPI showed higher values for hardness than that of the control-I. In general, hardness value of bologna with 3.0% protein additives were significantly ($P < 0.05$) higher than with 1.5% of their counterpart. Gnanasambandam & Zayas (1992) reported that firmness of soy protein and wheat protein in frankfurters increased when compared with control, which had same level of added water. Chin et al. (1999) reported that hardness of low-fat bologna with SPI depended upon the level of SPI in the formulation; formulations with 0 and 2.2% SPI had similar hardness values but the addition of 4.4% SPI decreased hardness ($P < 0.05$).

Adhesiveness values of protein added LFPB did not significantly differ from the control-I. In the same way, cohesiveness of the LFPB with protein extenders was not changed from the control-I. However, there were no significant differences in cohesiveness ($P > 0.05$) between control-II and bologna with Kabuli CPI, Desi CPI and SPI at 3.0% protein concentration.

Table 4.2.19 Effect of different protein binders on textural properties of cooked bologna

Treatment			Allo-Kramer	Texture profile analysis				
Binder		Binder level	Shear	Hardness	Adhesiveness	Cohesiveness[2]	Springiness	Chewiness
		(%)	(N/g)	(N)	(N)	-	(%)	N mm
Control	I	0	12.4 ^{e} ± 0.3	86.7 ^{e} ±5.3	-0.81 ^{b} ± 0.02	0.41 ^{bcd} ± 0.01	82.1 ^{a} ± 2.1	355.1 ^{e} ± 39.9
	II	0	23.1 ^{a} ± 1.0	191.1 ^{a} ± 5.2	-1.17 ^{c} ± 0.25	0.48 ^{a} ± 0.03	82.5 ^{a} ± 2.3	920.3 ^{a} ± 25.0
CPI	Kabuli	1.5	12.5 ^{e} ± 0.4	83.2 ^{e} ± 6.2	-0.70 ^{a} ± 0.10	0.41 ^{bcd} ± 0.06	80.4 ^{a} ± 3.6	434.7 ^{d} ± 27.8
		3.0	13.3 ^{ed} ± 1.5	119.7 ^{cb} ± 3.9	-0.90 ^{b} ± 0.00	0.44 ^{ab} ± 0.04	79.5 ^{a} ± 2.6	451.3 ^{d} ± 33.4
	Desi	1.5	13.3 ^{cde} ± 0.4	106.7 ^{d} ± 4.6	-0.87 ^{b} ± 0.06	0.37 ^{cd} ± 0.04	80.7 ^{a} ± 2.9	453.9 ^{d} ± 30.1
		3.0	14.7 ^{c} ± 0.1	117.7 ^{c} ± 4.2	-0.90 ^{b} ± 0.17	0.43 ^{abc} ± 0.04	80.6 ^{a} ± 1.3	463.2 ^{d} ± 13.8
SPI		1.5	14.6 ^{cd} ± 1.2	88.2 ^{e} ±7.3	-0.77 ^{b} ± 0.12	0.41 ^{bcd} ± 0.04	81.5 ^{a} ± 1.5	536.2 ^{c} ± 18.4
		3.0	16.9 ^{b} ± 0.8	127.5 ^{b} ± 2.2	-0.93 ^{b} ± 0.25	0.46 ^{ab} ± 0.06	81.4 ^{a} ± 2.7	643.2 ^{b} ± 18.9
PPI		1.5	13.5 ^{cde} ± 0.8	91.9 ^{e} ± 8.6	-0.77 ^{b} ± 0.12	0.36 ^{d} ± 0.01	81.4 ^{a} ± 1.3	327.4 ^{e} ± 16.8
		3.0	14.5 ^{cd} ± 1.2	102.7 ^{d} ± 2.1	-0.85 ^{b} ± 0.13	0.41 ^{bcd} ± 0.01	81.0 ^{a} ± 2.5	440.7 ^{d} ± 37.9

$^{a-e}$ Means within the same column with the same letter are not significantly different (P > 0.05)
[1] Non meat protein : CPI-chickpea protein isolate, SPI-soy protein isolates, PPI- pea protein isolates
[2] This value is dimensionless.

Springiness was not affected ($P > 0.05$) by the type of proteins or even meat protein level. Control-II samples apparently will require greater force to chew than the control-I and all other formulations. Hence the protein added bologna samples had significantly ($P < 0.05$) higher chewiness scores than that of the control-I except samples containing 1.5% PPI. LFPB samples with SPI seem to be chewier than the LFPB samples with CPI and PPI. Addition of CPI (except (3.0% Desi CPI) and 1.5% PPI had similar ($P < 0.05$) Kramer shear values (N/g) with compared to the control-I. Similar to the TPA-hardness, it is also evident that the SPI at 3.0% turned out the second firmest structure among the treatments. Chin et al. (1999) reported that low fat bologna with konjac blends had slightly higher ($P < 0.05$) Kramer shear values but values decreased as SPI in treatment increased.

As expected, the highest shear stress (kPa) and torsion rigidity values were noted for the control-II (Table 4.2.20). Due to high meat in the formulation, control-II (14.1% P) gave the "toughest" product (Figure 4.2.4) which was significantly different from the high water, lower meat control bologna (11.1% P). The second highest shear stress values were obtained for samples containing 3.0% SPI. Except for that formulation, other LFPB containing legume proteins did not significantly differ from the control-I. True shear strain at failure was not affected ($P > 0.05$) by any treatment factors with the exception of 3.0% Desi CPI. Since changes of shear strain among treatments were very minor torsion rigidity values were parallel to the shear stress scores (Table 4.2.20).

According to Figure 4.2.4, compared to the control, textural structure of LFPB with PPI and SPI at 3.0% level changed toward more "brittle" while texture of bologna with 3.0% Desi CPI changed from "rubbery" to more "mushy". Total opposite structural changes were given by the Kabuli CPI.

Table 4.2.20 Effect of different protein binders on torsion parameters of low fat bologna

Treatment			Torsion shear values		
Binder		Binder level (%)	Stress (kPa)	Strain[2]	Rigidity (kPa)
Control	I	0	33.28 ^{cd} ±1.84	1.45 ^{a} ±0.18	23.13 ^{cd} ±1.46
	II	0	60.52 ^{a} ±1.93	1.43 ^{a} ±0.06	42.48 ^{a} ±1.27
CPI	Kabuli	1.5	25.12 ^{e} ±0.85	1.28 ^{ab} ±0.07	19.73 ^{d} ±1.67
		3.0	30.41 ^{cde} ±1.33	1.45 ^{a} ±0.08	21.14 ^{d} ±0.56
	Desi	1.5	29.98 ^{de} ±1.49	1.38 ^{ab} ±0.13	21.71 ^{d} ±1.25
		3.0	29.62 ^{de} ±0.28	1.22 ^{b} ±0.08	22.92 ^{cd} ±0.65
SPI		1.5	35.88 ^{cd} ±1.46	1.45 ^{a} ±0.11	24.44 ^{cd} ±1.74
		3.0	44.26 ^{b} ±1.86	1.43 ^{a} ± 0.21	30.62 ^{b} ±1.12
PPI		1.5	31.31 ^{cde} ±1.65	1.41 ^{a} ±0.07	22.15 ^{cd} ±2.37
		3.0	38.06 ^{cb} ±1.18	1.42 ^{a} ±0.11	26.79 ^{cb} ±1.24

$^{a-e}$ Means within the same column with the same letter are not significantly different (P > 0.05)
[1] Non meat protein : CPI-chickpea protein isolate, SPI-soy protein isolates, PPI- pea protein isolates
[2] Shear strain is dimensionless.

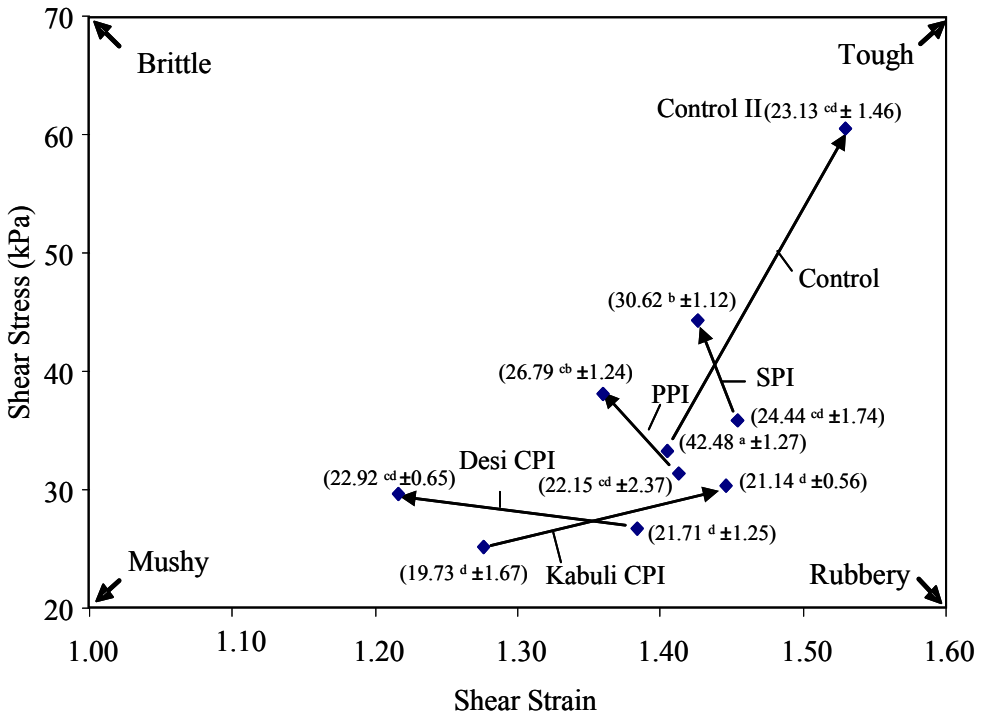

Figure 4.2.4 Torsion texture map of low-fat bologna containing different protein isolates. Arrows point from the 1.5% to the corresponding 3.0% of substitute plant protein level. Corresponding torsion rigidity values (shear stress/shear strain) (kPa) are given in parentheses. $^{a-d}$ Means of torsion rigidity with the same letter are not significantly different (P > 0.05).

So far, replacement of water with 1.5 or 3.0% protein from CPI/PPI in LFPB resulted in similar or stronger texture properties than the control-I. SPI generally gave stronger textures than the other two legume counterparts in the LFPB. Control-II had superior texture parameters to all other treatments. As determined in study 1, legume proteins (CPI, SPI and PPI) had denaturation temperature >90°C (Figure 4.1.11 and Table 4.1.18). LFPB were cooked only up to 72°C, hence, legume proteins in a meat system (bologna) would not be fully functional as gelling ingredients. This may be the reason they had a more brittle structure when compared to the control-II. Therefore, legume protein may act more as a filler in the LFPB formulations.

Table 4.2.21 shows the correlations among the textural parameters of LFPB with different protein additives. Shear strain was strongly correlated with shear stress (r = 0.96, P<0.001). Shear stress, which is the force required for fracture, also showed strong positive correlations with TPA-hardness (r = 0.78, P<0.001) and A-K shear force (r = 0.87, P<0.001) which measure the force needed to attain a given deformation. Torsion rigidity, which is an indication of stiffness, showed fairly strong positive correlations with hardness, chewiness and A-K shear forces (r = 0.81, 0.85 and 0.87, respectively, P<0.001) and a negative correlation with adhesiveness (r = 0.61, P<0.001). Further TPA hardness had strong positive correlations with cohesiveness, chewiness and A-K shear values and a strong negative relationship with adhesiveness (Table 4.2.21). Adhesiveness was significantly and negatively correlated with all other texture scores except shear strain. Springiness measures recovery after a specimen is compressed, and was not correlated to A-K shear values or torsion parameters.

Table 4.2.21 Correlation coefficients (r) of torsion rigidity, TPA and A-K shear force values of cooked protein-LFPB samples (combined data, n=27)

	2	3	4	5	6	7	8	9
1. Shear stress	0.36	0.96***	0.78***	-0.67***	0.52**	0.16	0.84***	0.87***
2. Shear strain	1	0.11	0.08	-0.31	0.28	0.29	0.13	0.16
3. Torsion rigidity		1	0.81***	-0.61***	0.44*	0.09	0.85***	0.87***
4. Hardness			1	-0.70***	0.58***	0.18	0.88***	0.90***
5. Adhesiveness				1	-0.55**	-0.48**	-0.63***	-0.58***
6. Cohesiveness					1	0.42*	0.57**	0.49**
7. Springiness						1	0.09	0.08
8. Chewiness							1	0.93***
9. A-K shear								1

*, **, *** = Significant at p <0.05, 0.01 and 0.001, respectively.
1 – 3 Torsion shear test values; 4 – 8 TPA parameters; 9 Allo-Kramer shears

4.2.2.7 Sensory properties

A 14-member sensory panel evaluated bologna slices (total seven treatments; control-I and bologna with 1.5/3.0% Kabuli and Desi isolates, 1.5% SPI and 1.5% PPI) from each replication. There were no differences ($P<0.05$) between treatments for cohesiveness and overall flavour intensities. However, there were differences ($P<0.05$) among treatments for juiciness, firmness, graininess, flavour desirability and foreign flavour intensities (Table 4.2.22 and Figure 4.2.5).

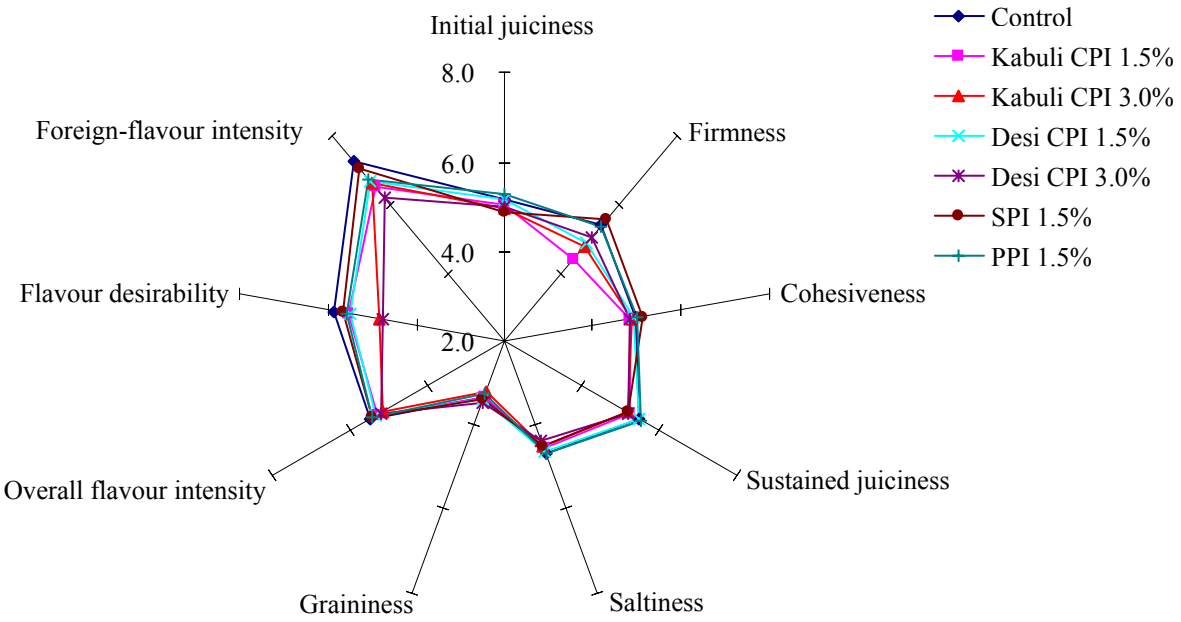

Figure 4.2.5 Sensory evaluation of LFPB formulated with Kabuli, Desi, pea and soy proteins at 1.5% and 3.0% levels. Initial and sustained juiciness: 8 (extremely moist) to 1 (extremely dry); firmness: 8 (extremely firm) to 1 (extremely soft); cohesiveness: 8 (extremely cohesive) to 1 (extremely brittle); saltiness: 6 (extremely salty) to 1 (not detectable); graininess 6 (extremely grainy) to 1 (not detectable); overall flavour intensity: 8 (extremely intense) to 0 (extremely band); flavour desirability: 8 (extremely desirable) to 1 (extremely undesirable); foreign flavour: 8 (no foreign flavour) to 1 (extremely intense foreign flavour)

Table 4.2.22 Effect of different protein binders on sensory properties[2] of low-fat pork bologna

Treatment		Initial juiciness	Firmness	Cohesiveness	Sustained juiciness	Graininess[3]
Binder[1]	Level (%)					
Control I	0	5.17 [ab] ±0.75	5.38 [ab] ± 0.95	4.95 [a] ±1.14	5.49 [a] ±0.67	1.34 [ab] ±0.55
CPI Kabuli	1.5	5.05 [bc] ±0.92	4.59 [d] ± 1.15	4.85 [a] ±1.09	5.23 [bc] ±0.84	1.51 [ab] ±0.68
	3.0	4.98 [c] ±0.73	5.29 [abc] ± 1.03	4.86 [a] ±1.05	5.17 [c] ±0.67	1.22 [b] ±0.53
Desi	1.5	5.17 [ab] ±0.97	4.85 [cd] ± 0.87	4.93 [a] ±0.83	5.43 [ab] ±0.77	1.37 [ab] ±0.68
	3.0	5.00 [bc] ±0.85	4.99 [bcd] ± 0.89	4.83 [a] ±0.96	5.20 [c] ±0.75	1.46 [a] ±0.76
SPI	1.5	4.88 [c] ±0.80	5.51 [a] ± 0.83	5.12 [a] ±0.92	5.17 [c] ±0.63	1.37 [ab] ±0.59
PPI	1.5	5.30 [a] ±0.72	4.80 [d] ± 0.67	5.02 [a] ±0.97	5.52 [a] ±0.67	1.27 [a] ±0.55

[1] Non meat protein : CPI-chickpea protein isolate, SPI-soy protein isolates, PPI- pea protein isolates

[a-d] Means within the same column with the same letter are not significantly different ($P > 0.05$)

[2] Highest possible score = 8 (extremely juicy, firm, cohesive, salty, intense, desirable, no foreign flavour); [2]lowest possible score = 1 (extremely dry, soft, brittle, bland, undesirable, intense) except for graininess or saltiness

[3] Highest possible score = 6 (extremely grainy or saltiness); [3] lowest possible score = 1 (no detectable graininess or saltiness)

Table 4.2.22 continued ….

Treatment			Saltiness[3]	Overall flavour intensity	Flavour desirability	Lack of foreign flavour intensity
Binder[1]		Level (%)				
Control	I	0	2.66 [a] ± 0.95	5.46 [a] ±0.79	5.86 [a] ±0.96	7.22 [a] ±0.98
CPI	Kabuli	1.5	2.54 [a] ± 1.15	5.27 [a] ±0.72	5.35 [a] ±1.14	6.49 [ab] ±1.64
		3.0	2.51 [a] ± 1.03	5.12 [a] ±1.27	4.84 [b] ±1.20	6.60 [ab] ±1.42
	Desi	1.5	2.65 [a] ± 0.87	5.32 [a] ±0.92	5.34 [a] ±1.06	6.67 [ab] ±1.48
		3.0	2.40 [a] ± 0.89	5.20 [a] ±1.17	4.76 [b] ±1.11	6.17 [b] ±1.93
SPI		1.5	2.53 [a] ± 0.83	5.40 [a] ±0.76	5.66 [a] ±0.66	7.01 [ab] ±1.01
PPI		1.5	2.66 [a] ± 0.67	5.39 [a] ±0.72	5.57 [a] ±1.12	6.68 [ab] ±1.53

[1] Non meat protein : CPI-chickpea protein isolate, SPI-soy protein isolates, PPI- pea protein isolates
[a-d] Means within the same column with the same letter are not significantly different (P > 0.05)
[2] Highest possible score = 8 (extremely juicy, firm, cohesive, salty, intense, desirable, no foreign flavour); [2]lowest possible score = 1 (extremely dry, soft, brittle, bland, undesirable, intense) except for graininess or saltiness
[3] Highest possible score = 6 (extremely grainy or saltiness); [3] lowest possible score = 1 (no detectable graininess or saltiness)

Only SPI and PPI at 1.5% addition levels in LFPB were used for the sensory study, in order to compare to commercial practices and reduce the number of samples given to the panelists. Perception of initial juiciness was not significantly ($P<0.05$) affected by addition of 1.5% Kabuli CPI, PPI and Desi CPI (both 1.5 and 3.0%) when compared to the control-I bologna. Bologna with 3.0% CPI and 1.5% SPI addition were slightly less juicy than the control. Sensory panel scores for sustained juiciness (determined during mastication) followed similar trends as the results for the initial juiciness; bologna with 1.5% Desi CPI and PPI had similar overall juiciness as the control-I. Adding Kabuli CPI (both 1.5 & 3.0% addition levels) and 1.5% SPI or Desi CPI to bologna reduced juiciness scores compared to that of the control-I. Chickpea proteins at 1.5 and 3.0% SPI exhibited the same scores for firmness as the control-I. However, LFPB having 1.5% CPI and PPI were perceived to be softer than the control-I with no binder. Generally sensory firmness values were similar to that obtained for TPA.

Shand (2000) observed that low-fat bologna with SPI (1.0%) and the nonadditive control were perceived to be softer ($P < 0.05$) than all other treatments. There was only a trivial difference for graininess and saltiness among treatments. Increased level of chickpea proteins in LFPB formulations decreased the perception of flavour desirability significantly ($P < 0.05$). This may be due to the foreign flavour introduced by the CPI at high level (i.e. 3.0% protein). For instance, bologna with Desi CPI at 3.0% protein addition had the highest score for lack of foreign flavour intensity (6.17) (ie moderately weak foreign flavour) and this formulation had the lowest perception of flavour desirability. Overall, flavour properties of CPI containing LFPB were as good as other legumes.

With few exceptions, significant correlation between instrumental texture properties and the sensory scores were not observed (Table 4.2.23). However there were strong and positive correlations between torsion stress and strain with sensory firmness ($r = 0.66$, $P < 0.001$; $r = 0.62$, $P < 0.01$, respectively). A significant ($P < 0.001$) inverse relationship ($r = -0.72$) was also observed between TPA hardness and sensory flavour desirability.

Table 4.2.23 Correlation coefficients (r) of the instrumental texture studies and sensory data (combined data, n=21)

	10	11	12	13	14	15	16	17	18
1. Torsion shear	-0.07	0.66***	0.23	0.09	0.02	-0.02	0.20	0.33	0.32
2. Torsion strain	0.15	0.62**	0.37	0.29	0.13	-0.57**	-0.04	0.51*	0.51*
3. Torsion rigidity	-0.18	0.38	0.05	-0.09	-0.09	0.29	0.25	0.14	0.09
4. TPA-Hardness	-0.15	0.10	-0.14	-0.20	-0.18	0.07	-0.33	-0.72***	-0.42
5. TPA-Adhesiveness	0.12	-0.23	0.00	0.18	0.32	0.03	0.45*	0.26	0.08
6. TPA-Cohesiveness	-0.17	0.39	-0.24	-0.18	-0.03	-0.05	-0.32	-0.19	0.13
7. TPA-Springiness	-0.02	0.18	-0.27	-0.05	-0.38	-0.10	-0.33	-0.03	0.22
8. TPA-Chewiness	-0.46*	0.28	0.09	-0.47*	-0.32	0.26	-0.18	-0.29	-0.16
9. A-K shear	-0.19	0.21	0.30	-0.14	-0.22	0.19	0.06	-0.34	0.42
10. Initial juiciness	1	-0.11	0.01	0.90***	0.23	-0.24	-0.11	0.30	0.27
11. Firmness		1	0.38	-0.01	0.05	-0.12	0.03	0.28	0.44*
12. Cohesiveness			1	0.11	-0.10	-0.13	-0.07	0.24	0.05
13. Sustained juiciness				1	0.43	-0.31	0.12	0.42	0.34
14. Saltiness					1	-0.27	0.65**	0.45*	0.26
15. Graininess						1	0.10	-0.14	-0.12
16. Overall flavour intensity							1	0.38	0.15
17. Flavour desirability								1	0.79***
18. Foreign flavour intensity									1

*, **, *** = Significant at P < 0.05, 0.01 and 0.001, respectively.

As expected, initial juiciness was highly correlated with sustained juiciness, similarly; saltiness was significantly correlated with overall flavour intensity and flavour desirability; foreign flavour was highly correlated ($r = 0.79$, $P < 0.001$) with flavour desirability of LFPB with various protein binders.

4.2.2.8 Summary and conclusions

In summary, the results indicate that using CPI in LFPB did not cause any detrimental effect on the meat system. Additions of Kabuli and Desi CPI improved batter characteristics by increasing water holding properties and decreasing cooking loss. The temperature of batters containing protein additives showed an increase when compared to the control-I. A positive correlation between water holding capacity and viscosity of meat batters was also observed. Three different texture tests including torsion shear test, TPA and Allo-Kramer shear force were conducted in order to understand textural behavior of LFPB. It was confirmed that product having the highest meat protein content (control-II) was the toughest product. However, products containing 3.0% CPI, PPI and SPI were harder than those containing their 1.5% counterparts or the control-I (having same level of meat proteins). Generally, addition of plant protein ingredients in LFPB did not change the L* and a* values but they significantly increased the yellowness (b*) of the products. CPI, PPI and SPI at 1.5% addition level in LFPB did not alter flavour properties of the products. Therefore CPI is a potential source of non-meat protein for emulsion type meat products.

4.2.3 Chickpea starch as an extender in low-fat pork bologna

4.2.3.1 Starch properties

Table 4.2.24 shows the results of the chemical analysis of the four different starch extenders. Potato starch was slightly lower in starch and higher in moisture content than chickpea and pea starch. Protein, fat and ash contents were very small in all starch samples.

Table 4.2.24 Proximate composition[a] of starches

Binder	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Starch (%)
Kabuli	8.3 ± 0.06	0.49 ± 0.25	0.09 ± 0.05	0.10 ± 0.03	98.0 ± 0.80
Desi	9.2 ± 0.04	0.80 ± 0.04	0.07 ± 0.01	0.12 ± 0.00	96.8 ± 0.95
Potato	12.2 ± 0.01	0.00 ± 0.00	0.05 ± 0.00	0.15 ± 0.07	91.0 ± 1.20
Pea	10.5 ± 0.04	0.54 ± 0.05	0.52 ± 0.64	0.14 ± 0.06	97.6 ± 0.98

[a] Mean (± SD) of duplicate determinations (reported on an as is basis, starch values are moisture free basis)

4.2.3.2 Raw batter properties

Temperature of the meat batter was checked during the each stage of the bologna preparation. Average temperature of meat at the beginning was between -1 and 0°C. Average temperature of meat batter after mixing with ingredients/ice water, and just before the vacuum step, was 7.1-8.3°C. Temperature control is very important during emulsifying. The batter must warm slightly (through friction) enough to allow microscopic fat particles to be encapsulated by the meat proteins but not over-chopped to liquefy too much of the lipid. Batter viscosity and temperatures measured immediately after stuffing are tabulated in Table 4.2.25. Mixing starch into the batters lead to a significant ($P < 0.05$) increase in batter temperature by 2-3°C, except Desi starch at 1.0% addition which had no effect. Formulations with 2.0% Kabuli and Desi starch showed significantly ($P < 0.05$) higher temperature when compared with those with 1.0% starch addition. The increased temperatures are likely due to the 1% less ice water in those formulations and the temperature of the ingredients which were at room temperature.

Starch addition to the LFPB formulations resulted in lower ($P < 0.05$) batter viscosity than found for the control (Table 4.2.13). However, Shand (2000) formulated potato starch (4%) in ultra-low fat bologna (>1% fat) and found that starch increased the viscosity of the raw batter. Similarly, a study conducted by Aktas & Genceelep (2006) for modified/unmodified corn and potato starch in bologna-type sausage showed starch modification lead to a statistically significant increase in raw batter viscosity, although the effect of type of starch and type of starch × starch modification interactions were non-significant.

Table 4.2.25 Effect of starch binders on apparent viscosity of raw low-fat batters and batter temperature right after stuffing

Treatment			Viscosity	Batter temperature
Starch binder		Level (%)	cps × 10^5	°C
Control		0	2.55 ^{a} ± 0.03	8.3 ^{d} ± 0.3
Chickpea	Kabuli	1.0	2.32 ^{b} ± 0.02	10.7 ^{b} ± 0.6
		2.0	2.22 ^{bcd} ± 0.02	12.1 ^{a} ± 1.4
	Desi	1.0	2.27 ^{bc} ± 0.07	8.3 ^{d} ± 0.3
		2.0	2.13 ^{bcde} ± 0.05	10.5 ^{c} ± 0.4
Potato		1.0	2.12 ^{ced} ± 0.13	12.1 ^{a} ± 0.9
		2.0	2.07 ^{ed} ± 0.14	12.2 ^{a} ± 1.1
Pea		1.0	1.99 ^{e} ± 0.22	11.5 ^{ab} ± 1.4
		2.0	2.14 ^{cebd} ± 0.18	11.9 ^{a} ± 1.1

$^{a-e}$ Means (±SD) within the same column with the same letter are not significantly different ($P > 0.05$)
cps – centipoise unit

4.2.3.3 Chemical composition and pH of the cooked bologna

Except moisture content of LFPB with 2.0% potato starch, no differences ($P < 0.05$) was found in moisture, protein and pH of treatments in comparison with the control (Table 4.2.26). The chemical composition of our LFPB was very similar to previous low-fat bologna studies (Shand, 2000; Chin et al., 2000).

Table 4.2.26 Proximate analysis of LFPB formulated with various starch binders

Treatment			Moisture	Protein	pH
Starch binder		Level (%)	(%)	(%)	
Control		0	78.9 [a] ± 0.65	14.4 [ab] ± 0.53	6.14 [a] ± 0.07
Chickpea	Kabuli	1.0	77.8 [abc] ± 1.17	14.0 [ab] ± 0.46	6.24 [a] ± 0.03
		2.0	77.5 [abc] ± 0.84	13.7 [ab] ± 0.66	6.20 [a] ± 0.17
	Desi	1.0	77.5 [abc] ± 1.85	14.3 [ab] ± 1.41	6.32 [a] ± 0.14
		2.0	77.1 [abc] ± 0.87	14.1 [ab] ± 0.44	6.21 [a] ± 0.00
Potato		1.0	78.6 [ab] ± 0.44	13.5 [b] ± 0.56	6.23 [a] ± 0.12
		2.0	76.5 [c] ± 2.18	14.6 [a] ± 0.85	6.24 [a] ± 0.06
Pea		1.0	78.8 [abc] ± 1.20	13.8 [ab] ± 0.81	6.29 [a] ± 0.24
		2.0	78.3 [ab] ± 1.65	13.6 [b] ± 1.36	6.31 [a] ± 0.21

[a-d] Means (± SD) within the same column with the same letter are not significantly different at $P < 0.05$

4.2.3.4 Cook yield, expressible moisture and purge losses

Cook yields of LFPB were significantly improved ($P < 0.05$) by the addition of the different starches (Table 4.2.27) with exception of 1.0% pea starch treatment. Bologna with Kabuli starch and potato starch at 2.0% showed the highest cook yield of 95.5 and 95.9% respectively, which were not significantly different ($P > 0.05$) between each other. However, bologna with pea starch and Desi starch at 2.0% was less effective with regards to cook yield than the potato and Kabuli starch counterparts. As a general trend, bologna with increased level of added starch (i.e. 2.0%) had higher cook yields than 1.0% addition of starch in the formulation of low-fat pork bologna. This was in agreement with the reduction of cook loss of starch/meat complex by corn/rice/pea mung bean starches (Li & Yeh, 2003), reduced-fat turkey batter by modified potato and tapioca starches (Hachmeister & Herald, 1998), and bologna by waxy corn starch (Carballo, Barreto, & Jimenez Colmenero, 1995). However, Shand (2000) and Yang et al. (2001) reported that cook yield with addition of starch in low-fat bologna and frankfurters, respectively, did not significantly differ from the control.

Table 4.2.27 Effect on starch binders on cook yield, purge losses and expressible moisture of cooked bologna

Treatment			Cook yield	Purge losses	Expressible moisture (%)
Binder		Level (%)	(%)	(%)	
Control		0	91.63 [c] ± 0.84	2.83 [a] ± 0.03	19.31[a] ± 2.67
Chickpea	Kabuli	1.0	94.35 [b] ± 0.69	2.01 [b] ± 0.16	14.70 [d] ± 3.78
		2.0	95.51 [a] ± 0.75	1.99 [b] ± 0.40	12.32 [f] ± 1.93
	Desi	1.0	93.80 [b] ± 0.46	2.39 [ab] ± 0.03	15.61 [c] ± 1.73
		2.0	94.45 [b] ± 0.67	1.70 [b] ± 0.14	12.75 [ef] ± 0.90
Potato		1.0	93.95 [b] ± 0.50	2.35 [ab] ± 0.02	17.05 [b] ± 1.47
		2.0	95.85 [a] ± 0.90	1.70 [b] ± 0.49	16.53 [b] ± 1.06
Pea		1.0	92.17 [c] ± 0.84	2.25 [ab] ± 0.77	16.41 [e] ± 1.62
		2.0	94.15 [b] ± 0.65	2.21 [ab] ± 1.03	14.74 [ef] ± 1.95

[a-d] Means (±SD) within the same column with the same letter are not significantly different (P > 0.05)

Expressible moisture (EM) showed differences (P < 0.05) between control bologna and the others (Table 4.2.27). Treatments with 1.0% Kabuli, 2.0% Kabuli, Desi and potato starches had significantly (P < 0.05) lower purge losses than the control. EM values of LFPB containing 2.0% Kabuli or Desi chickpea starch level tended to be lower than their 1.0% counterpart indicating that water holding capacity might be greater with 2.0% chickpea starch levels rather than the 1.0% level. Further, bologna with chickpea starch had lower EM values than with potato starch. A similar relationship was found for purge losses (Table 4.2.27). The control bologna with a high amount of water and no binders had significantly higher (P < 0.05) purge loss (2.8%) than that of the bologna with Kabuli chickpea starch (both 1.0 and 2.0%), Desi starch 2.0% and potato starch 2.0%. The decrease in purge was probably due to an increase in starch gelatinization during the cooking proccess. The beneficial effect of starch on reducing purge loss was similar to that reported by Yang, Keeton, Beilken, & Trout (2001). They found that 4% modified waxy maize starch in low fat frankfurters had reduced purge by 7.2% as compared with the control. Shand (2000) also found that ultra-low-fat pork bologna with 4% potato

starch had significantly lower purge loss than the control. However, Beggs et al. (1997) reported the usage of modified corn starch in twenty different formulations of reduced-fat turkey frankfurters and they did not observe significant differences of purge losses among the treatments.

The Pearson correlation coefficients of raw batter properties, cooking yield, EM and purge loss are shown in Table 4.2.28. Emulsion temperature had a small and positive correlation (r = 0.49) with cooking yield. As anticipated, formulations which had high water holding properties (low EM/purge loss) showed good relationship with cooking yield of LFPB. Correlation between EM and purge was significant (r = 0.45).

Table 4.2.28 Correlation coefficients (r) among raw batter properties, cooking yield, EM and purge loss of cooked LFPB (combined data, n=27)

	Batter temperature	Cooking yield	EM	Purge loss
Viscosity	-0.56**	-0.27	0.26	0.14
Batter temperature	1	0.49**	-0.26	-0.22
Cooking yield		1	-0.63***	-0.49*
EM			1	0.45*
Purge loss				1

*, **, *** = Significant at P < 0.05, 0.01 and 0.001, respectively.

4.2.3.5 CIE colour

There were no significant differences (P > 0.05) in objective colour measurements between treatments for L* and a* values (Table 4.2.29). With the b* values, there were significant differences among bologna with 2.0% Desi starch, potato starch and pea starch (P < 0.05) but these were small (ranging from 11.59 to 12.35) and were not meaningful. The effect of starch on the finished product was consistent with published findings. Shand (2000) reported that addition of 4% potato starch in low-fat pork bologna had no detrimental effect on colour of cooked bologna when compared with the control. Beggs et al. (1997) showed that level of

modified corn starch affected b* values but not L* or a* values in turkey frankfurters. Furthermore, starch and fiber additives had only minor effects on the colour of low-fat bologna (Claus & Hunt, 1991).

Table 4.2.29 Effect of different starch binders on colour of cooked low fat bologna

Treatment			CIE colour[1]		
Binder		Level (%)	L*	a*	b*
Control		0	68.60 [a] ± 0.28	18.37 [a] ± 0.13	11.74 [ab] ± 0.10
Chickpea	Kabuli	1.0	68.21 [a] ± 0.23	18.42 [a] ± 0.31	11.75 [ab] ± 0.17
		2.0	69.25 [a] ± 0.20	17.96 [a] ± 0.14	12.03 [ab] ± 0.13
	Desi	1.0	68.44 [a] ± 0.20	18.53 [a] ± 0.12	11.77 [ab] ± 0.08
		2.0	68.71 [a] ± 0.16	18.64 [a] ± 0.10	12.35 [a] ± 0.04
Potato		1.0	68.59 [a] ± 0.20	18.59 [a] ± 0.15	12.03 [ab] ± 0.06
		2.0	68.95 [a] ± 0.19	18.53 [a] ± 0.10	12.32 [a] ± 0.10
Pea		1.0	68.55 [a] ± 0.16	18.29 [a] ± 0.09	11.59 [b] ± 0.08
		2.0	68.16 [a] ± 0.58	18.23 [a] ± 0.33	11.63 [b] ± 0.18

[a-d] Means within the same column with the same letter are not significantly different (P > 0.05)
[1] CIE colour: "L*" = lightness; "a*" = redness; "b" = yellowness

4.2.3.6 TPA, Allo-Kramer shear and torsion analysis.

Incorporation of selected starches into the LFPB system significantly influenced most textural attributes (Table 4.2.30). The effect produced on the texture by different treatments could be attributed to the characteristics of the gel that is formed within the meat matrix by different botanical sources and level of starch. TPA-hardness showed significant differences (P< 0.05) among formulations. The lowest values for hardness were obtained for the control but these did not significantly differ from the 1.0% potato and pea starch containing samples. The highest TPA-hardness values were obtained for the bologna samples with Kabuli starch (at both levels) and 2.0% Desi starch. So chickpea samples had better texture (hardness) when compared to samples with potato or pea starch. Adhesiveness of all formulations with starch did not significantly (P>0.05) vary from the control, while addition of 2.0% Kabuli starch and

Table 4.2.30 Effect of different starch binders on instrumental textural properties of cooked low fat bologna

Treatment			Texture profile analysis					Allo-Kramer
Binder		Level (%)	Hardness (N)	Adhesiveness (N)	Cohesiveness[1] -	Springiness (%)	Chewiness N mm	Shear (N/g)
Control		0	95.4 ^{e} ± 0.9	-0.83 ^{ab} ± 0.06	0.36 ^{b} ± 0.01	75.5 ^{d} ± 1.0	441.4 ^{e} ± 6.5	11.4 ^{e} ± 0.5
Chickpea	Kabuli	1.0	121.9 ^{ab} ± 3.4	-0.83 ^{ab} ± 0.21	0.42 ^{ab} ± 0.03	82.4 ^{ab} ± 2.1	539.7 ^{bcd} ± 14.6	14.7 ^{bcd} ± 0.9
		2.0	131.1 ^{a} ± 1.8	-0.97 ^{b} ± 0.06	0.47 ^{a} ± 0.04	81.4 ^{ab} ± 0.3	605.7 ^{ab} ± 9.6	16.6 ^{a} ± 1.2
	Desi	1.0	112.0 ^{bcd} ± 3.3	-1.00 ^{b} ± 0.10	0.47 ^{a} ± 0.06	83.5 ^{a} ± 1.5	570.4 ^{abc} ± 18.5	14.0 ^{cd} ± 0.7
		2.0	123.4 ^{ab} ± 2.8	-0.90 ^{b} ± 0.10	0.44 ^{a} ± 0.06	80.5 ^{bc} ± 1.6	642.5 ^{a} ± 11.1	16.6 ^{a} ± 0.5
Potato		1.0	106.0 ^{cde} ± 6.4	-0.80 ^{ab} ± 0.10	0.43 ^{ab} ± 0.04	80.9 ^{ab} ± 2.3	481.3 ^{de} ± 17.0	14.9 ^{bc} ± 0.7
		2.0	113.8 ^{bc} ± 5.9	-0.67 ^{a} ± 0.15	0.40 ^{ab} ± 0.07	78.0 ^{cd} ± 1.2	465.2 ^{de} ± 19.9	15.8 ^{ab} ± 0.9
Pea		1.0	100.2 ^{de} ± 5.8	-0.93 ^{b} ± 0.12	0.43 ^{ab} ± 0.02	81.7 ^{ab} ± 1.4	413.1 ^{e} ± 17.1	13.5 ^{d} ± 0.8
		2.0	110.9 ^{bcd} ± 9.4	-0.87 ^{ab} ± 0.15	0.43 ^{ab} ± 0.05	80.6 ^{bc} ± 2.5	494.8 ^{cde} ± 15.2	14.3 ^{cd} ± 0.8

$^{a-d}$ Means within the same column with the same letter are not significantly different (P > 0.05)

[1] This value is dimensionless.

1.0/2.0% Desi starch resulted in higher ($P < 0.05$) cohesiveness than that of the control. Cohesiveness among other treatments was not significantly different. In the case of springiness, the control bologna had the lowest and 2.0% Desi samples had the highest springiness among the LFPB formulations. Springiness of LFPB containing 1.0% starch and Kabuli starch at 2.0% addition level was higher ($P < 0.05$) when compared with the rest of the treatments.

TPA chewiness of the control did not differ from bologna with pea or potato starch at both formulation levels although chickpea starches at 1.0 or 2.0% addition levels showed significantly ($P < 0.05$) higher values than the other treatments. As a general trend, textural properties of LFPB containing chickpea starch (both *var*.) were better than the control.

Allo-Kramer (A-K) shear values for bologna slices containing different starches from chickpea, pea and potato were significantly ($P < 0.05$) higher as compared to the control. There was a direct relationship ($P < 0.05$) to A-K shear as starch level increased (high level had high A-K shearing force), although pea starch containing samples did not seem to follow the pattern. Generally bologna with Kabuli starches at 2.0% were more firm, chewy, springy and adhesive than those with 2.0% potato or pea starch whereas cohesiveness and A-K shear force were similar in bologna with both starches. The 2.0% Kabuli starch containing LFPB had superior textural qualities to 2.0% pea starch bologna. According to our findings, 1.0% starch addition to the formulations did not reflect the botanical distinction of starches.

Differences ($P < 0.05$) were found in torsion shear values and torsion rigidity (shear stress/shear strain) in LFPB formulations with various starches (Table 4.2.31). The lowest value for shear stress was observed for 1.0% pea starch containing LFPB whereas the highest shear stress was noted for bologna with 2.0% Kabuli starch. Except for the 1.0% pea starch samples, shear stress of all other formulations had higher values than that of the control. There were no significant differences in shear strain among starch added formulations except value for bologna containing potato starch were higher than the control.

Table 4.2.31 Effect of different starch binders on torsion parameters of cooked low-fat bologna

Treatment			Torsion shear values		
Binder		Binder level (%)	Stress (kPa)	Strain[1]	Rigidity (kPa)
Control		0.0	30.27 ^{d} ± 0.22	1.31 ^{b} ± 0.00	23.11 ^{d} ± 0.17
Chickpea	Kabuli	1.0	37.50 ^{b} ± 0.46	1.41 ^{ab} ± 0.02	26.67 ^{bc} ± 0.65
		2.0	44.89 ^{a} ± 1.03	1.38 ^{ab} ± 0.11	32.73 ^{a} ± 2.45
	Desi	1.0	34.81 ^{c} ± 0.70	1.40 ^{ab} ± 0.10	25.01 ^{cd} ± 1.87
		2.0	37.96 ^{b} ± 1.42	1.38 ^{ab} ± 0.04	27.46 ^{b} ± 1.42
Potato		1.0	34.02 ^{c} ± 0.95	1.47 ^{a} ± 0.04	23.17 ^{d} ± 1.31
		2.0	37.66 ^{b} ± 1.09	1.35 ^{a} ± 0.12	27.92 ^{b} ± 1.66
Pea		1.0	27.98 ^{e} ± 1.27	1.39 ^{ab} ± 0.03	20.14 ^{d} ± 1.29
		2.0	33.76 ^{c} ± 1.09	1.46 ^{ab} ± 0.09	23.15 ^{e} ± 0.74

[a-e] Means within the same column with the same letter are not significantly different (P > 0.05)
[1] Shear strain is dimensionless

A texture map, which is a plot of shear stress vs shear strain, provides a graphical representation of product texture. A map illustrating the textural changes in the LFPB with different starches at 1.0 and 2.0% level is shown in Figure 4.2.6. Bologna exhibiting low shear stress are termed mushy if shear strain is low (a better descriptor for bologna would be "soft"), and rubbery if shear strain is high. Bologna with high shear stress values can be considered brittle if shear strain is low, and tough if shear strain is high. Formulations with 2.0% Kabuli, Desi and potato starch which have 2.0% less water, became slightly more brittle. On the contrary, texture of bologna with pea starch changed from "mushy" to "rubbery" when starch level increased from 1.0% to 2.0%. Desi starch containing formulations had the smallest changes of shear strain while potato starch showed the highest. For torsion rigidity values (shear stress/shear strain), bologna with 1.0 and 2.0% Kabuli, 2.0% Desi, potato and pea starches had a significantly higher value than that of the control. The control and bologna with pea and Desi starch (both 1.0 and 2.0%) or 1.0% potato starch showed the lowest torsion rigidity values, whereas, bologna with 2.0% Kabuli and potato starches were intermediate. Generally, LFPB

formulated with 2.0% Kabuli and Desi starch had better texture than those with pea and potato starch.

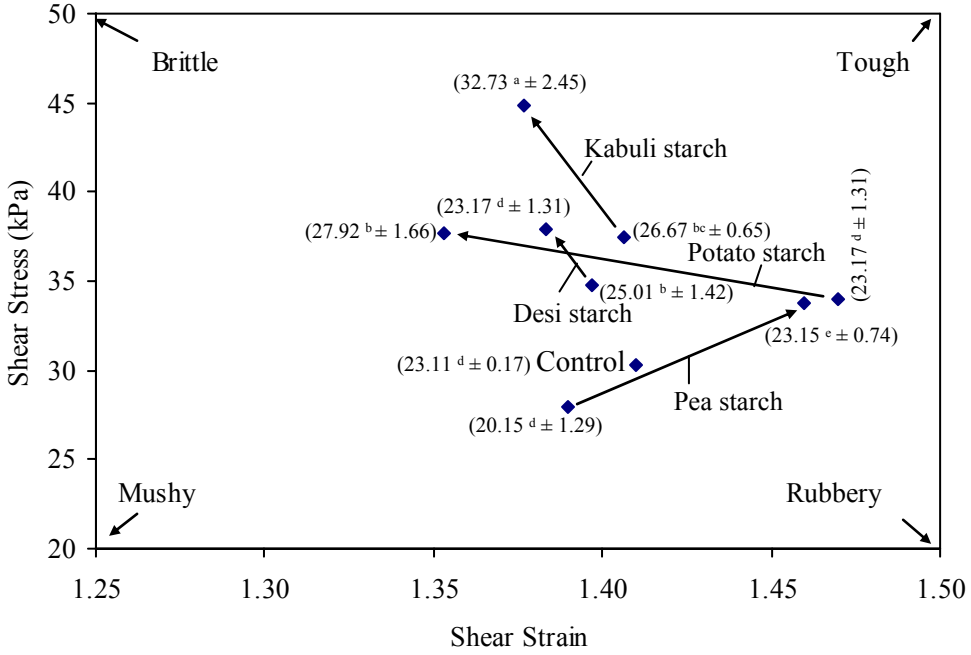

Figure 4.2.6 Torsion texture map of low fat bologna containing different starches. Arrows point from the 1.0% to the corresponding 2.0% of substitute starch level. Corresponding torsion rigidity values (shear stress/shear strain) (kPa) are given in parentheses. [a-b] Means of torsion rigidity with the same letter are not significantly different *(*P > 0.05).

Correlations among instrumental texture properties of starch-LFPB are shown in Table 4.2.32. Shear stress had a very strong correlation with torsion rigidity (r = 0.92, P<0.001) and strong positive correlation with A-K shear values (r = 0.63, P<0.01). There were only small but significant (P<0.05) correlations between shear strain and TPA springiness as well as with A-K shear force. Correlations occurred between hardness and torsion rigidity, chewiness and A-K shear values (r = 0.50, P<0.05; r = 0.80, P<0.001; r = 0.79, P<0.001, respectively). Similarly, very strong correlations (p<0.001) were established between cohesiveness and springiness (r = 0.77), and chewiness and A-K shear values (r = 0.66). Furthermore, there was a negative correlation between adhesiveness and cohesiveness (r = -0.60, P<0.001) as well as with springiness (r = -0.46, P<0.05).

Table 4.2.32 Correlation coefficients (*r*) of torsion rigidity, TPA and A-K shear force values of cooked starch-LFPB samples (combined data, n=27)

	2	3	4	5	6	7	8	9
1. Torsion stress	0.02	0.92***	0.16	0.03	-0.05	0.00	0.06	0.63**
2. Torsion strain	1	-0.36	0.16	-0.11	0.31	0.41*	0.21	0.44*
3. Torsion rigidity		1	0.50*	-0.39	0.20	0.05	0.24	0.40
4. Hardness			1	-0.26	0.27	0.29	0.80***	0.79***
5. Adhesiveness				1	-0.60***	-0.46*	-0.37	-0.14
6. Cohesiveness					1	0.77***	0.40*	0.45*
7. Springiness						1	0.36	0.38*
8. Chewiness							1	0.66***
9. A-K shear								1

*, **, *** = Significant at $p < 0.05$, 0.01 and 0.001, respectively.

4.2.3.7 Sensory properties

Results from sensory evaluation of LFPB with chickpea, pea and potato starch are represented in Figure 4.2.7 and Table 4.2.33. It is very clear that addition of starch had few effects on sensory properties. Initial juiciness and sustained juiciness of bologna formulations with added starch were not significantly different from the control.

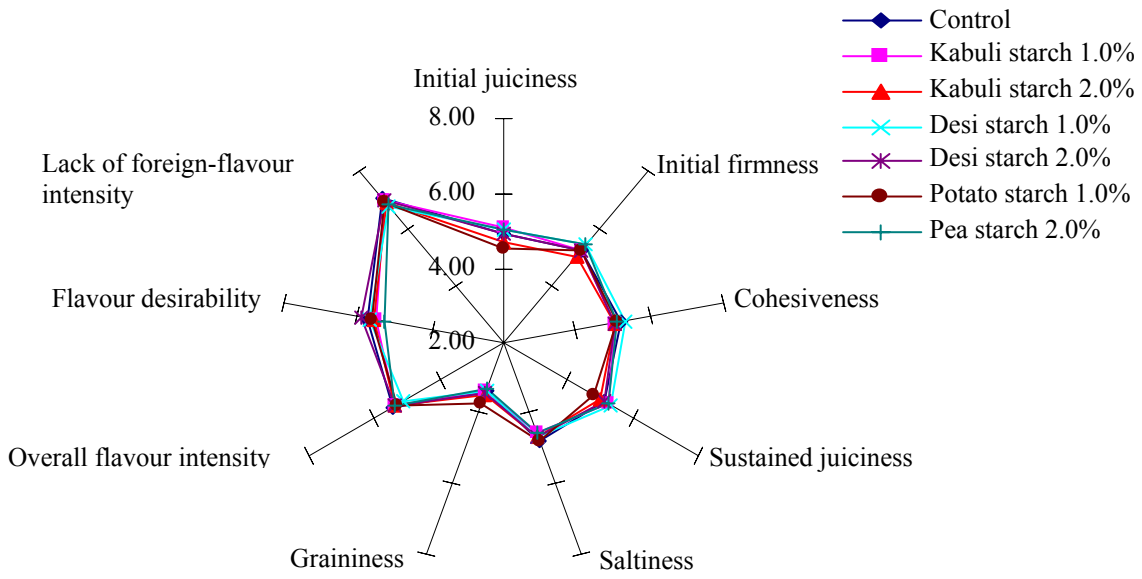

Figure 4.2.7 Sensory evaluation of LFPB formulated with Kabuli, Desi, pea and potato starch at 1.0% and 2.0% levels. Initial and sustained juiciness: 8 (extremely moist) to 1 (extremely dry); firmness: 8 (extremely firm) to 1 (extremely soft); cohesiveness: 8 (extremely cohesive) to 1 (extremely brittle); saltiness: 6 (extremely salty) to 1 (not detectable); graininess 6 (extremely grainy) to 1 (not detectable); overall flavour intensity: 8 (extremely intense) to 1 (extremely band); flavour desirability: 8 (extremely desirable) to 1 (extremely undesirable); foreign flavour: 8 (no foreign flavour) to 1 (extremely intense foreign flavour)

Table 4.2.33 Effect of different starch binders on sensory properties[1] of low-fat pork bologna

Treatment			Initial juiciness	Firmness	Cohesiveness	Sustained juiciness	Graininess[2]
Starch binder		Level (%)					
Control		0	4.90 ^{ab} ± 0.02	5.19 ^{a} ± 0.07	5.18 ^{a} ± 0.26	5.16 ^{ab} ± 0.06	1.36 ^{b} ± 0.18
Chickpea	Kabuli	1.0	5.06 ^{a} ± 0.43	5.26 ^{a} ± 0.14	5.04 ^{a} ± 0.04	5.24 ^{a} ± 0.17	1.39 ^{ab} ± 0.19
		2.0	4.71 ^{ab} ± 0.26	5.06 ^{a} ± 0.23	5.02 ^{a} ± 0.26	4.96 ^{ab} ± 0.29	1.49 ^{ab} ± 0.10
	Desi	1.0	5.05 ^{a} ± 0.13	5.43 ^{a} ± 0.17	5.30 ^{a} ± 0.27	5.30 ^{a} ± 0.21	1.35 ^{b} ± 0.15
		2.0	4.91 ^{ab} ± 0.32	5.28 ^{a} ± 0.29	5.04 ^{a} ± 0.27	5.13 ^{ab} ± 0.37	1.44 ^{ab} ± 0.24
Potato		2.0	4.54 ^{b} ± 0.04	5.21 ^{a} ± 0.11	5.11 ^{a} ± 0.09	4.83 ^{b} ± 0.10	1.76 ^{a} ± 0.19
Pea		2.0	5.00 ^{a} ± 0.15	5.41 ^{a} ± 0.26	5.09 ^{a} ± 0.26	5.28 ^{a} ± 0.13	1.31 ^{b} ± 0.12

$^{a-d}$ Means within the same column with the same letter are not significantly different (P>0.05)

[1] Highest possible score = 8 (extremely juicy, firm, cohesive, salty, intense, desirable, no foreign flavour); [1]lowest possible score = 1 (extremely dry, soft, brittle, bland, undesirable, intense) except for graininess or saltiness

[2] Highest possible score = 6 (extremely grainy or saltiness); [2] lowest possible score = 1 (no detectable graininess or saltiness)

Table 4.2.33 continued ….

Treatment		Saltiness[2]	Overall flavour intensity	Flavour desirability	Lack of foreign flavour intensity
Starch binder	Level (%)				
Control	0	2.82 [a] ± 0.05	5.41 [a] ± 0.42	5.70 [a] ± 0.42	7.05 [a] ± 0.12
Chickpea Kabuli	1.0	2.58 [b] ± 0 .19	5.38 [a] ± 0.62	5.49 [ab] ± 0.41	6.99 [a] ± 0.24
	2.0	2.68 [ab] ± 0.17	5.34 [a] ± 0.34	5.53 [ab] ± 0.34	6.91 [a] ± 0.20
Desi	1.0	2.68 [ab] ± 0.09	5.16 [a] ± 0.49	5.62 [ab] ± 0.58	6.74 [a] ± 0.18
	2.0	2.67 [ab] ± 0.01	5.36 [a] ± 0.04	5.85 [a] ± 0.02	7.00 [a] ± 0.23
Potato	2.0	2.82 [a] ± 0.11	5.29 [a] ± 0.29	5.58 [ab] ± 0.17	6.93 [a] ± 0.36
Pea	2.0	2.56 [b] ± 0.12	5.36 [a] ± 0.58	5.23 [b] ± 0.57	6.81 [a] ± 0.56

[a-d] Means within the same column with the same letter are not significantly different (P>0.05)

[1] Highest possible score = 8 (extremely juicy, firm, cohesive, salty, intense, desirable, no foreign flavour); [1]lowest possible score = 1 (extremely dry, soft, brittle, bland, undesirable, intense) except for graininess or saltiness

[2] Highest possible score = 6 (extremely grainy or saltiness); [2] lowest possible score = 1 (no detectable graininess or saltiness)

Samples containing 2.0% chickpea (Kabuli and Desi) starch had similar initial and sustained juiciness as the 1.0% counterpart. Moreover, 1.0% Kabuli and Desi starch treatments were more ($P < 0.05$) juicy than that with 2.0% potato starch. Unlike TPA findings, panelists didn't perceive any difference in hardness and cohesiveness between treatments ($P > 0.05$). Similarly there was no difference in flavour attributes (overall flavour intensity and foreign flavour intensity) among treatments. Saltiness of the samples containing 1.0% Kabuli starch and bologna with 2.0% pea starch had lower ($P < 0.05$) values than that of the non additive control and the 2.0% potato starch. There were significant ($P < 0.05$) but small differences in graininess between treatments and between treatments and the control (Table 4.2.21). However, the highest value for graininess was obtained for bologna with 2.0% potato starch.

Claus & Hunt (1991) reported that addition of wheat starch in 10 and 30% fat bologna did not significantly change the graininess from the control. However, all bologna containing dietary fiber were scored as having more graininess ($P < 0.05$) than the 30% fat and 10% fat control bologna (Claus & Hunt, 1991). Even though overall flavour intensities were not significantly different, flavour desirability differed slightly between treatments. The control and 2.0% Desi starch had slightly higher flavour desirability scores than that of the treatment with added pea starch (2.0%). Chin et al. (2000) reported that there were no overall differences ($P > 0.05$) in flavour and taste attributes between low-fat bologna combinations (konjac flour, carrageenan and starch 0.5 and 1.0%) and the control with 30% fat. In the present study, LFPB containing chickpea, potato and pea starch either at 1.0% or 2.0% was given an acceptable sensory texture and flavour scores which were similar to the control.

The Pearson correlation coefficient within and between sensory and instrumental attributes are presented in Table 4.2.34. There were few correlations found between sensory and instrumental textural attributes. The only meaningful relationship was a correlation between TPA-cohesiveness and sensory firmness ($r = 0.44$, $P<0.05$).

Table 4.2.34 Correlation coefficients (*r*) of the instrumental texture and sensory studies of starch-LFPB study (combined data, n=21)

	8	9	10	11	12	13	14	15	16
1. Torsion rigidity	-0.48*	0.08	-0.05	-0.35	-0.01	0.24	0.36	0.20	0.35
2. TPA-hardness	-0.16	0.06	-0.39	-0.20	-0.21	0.23	0.11	0.08	-0.04
3. TPA-adhesiveness	-0.21	-0.28	-0.07	-0.23	0.34	0.37	-0.20	-0.15	-0.08
4. TPA-cohesiveness	0.06	0.44*	0.15	0.11	-0.36	-0.37	-0.20	-0.02	0.03
5. TPA-springiness	0.28	0.31	0.07	0.30	-0.46*	-0.36	-0.30	-0.19	-0.24
6. TPA-chewiness	0.07	0.01	-0.15	0.12	-0.07	-0.13	0.03	0.29	-0.02
7. A-K shear	-0.34	-0.05	-0.31	-0.33	-0.13	0.34	0.02	0.07	0.00
8. Initial juiciness	1	0.27	0.38	0.86***	-0.26	-0.57**	-0.02	0.09	-0.13
9. Firmness		1	0.37	0.42	-0.36	-0.48	-0.16	0.08	0.20
10. Cohesiveness			1	0.54*	-0.13	-0.36	-0.18	0.13	0.35
11. Sustained juiciness				1	-0.41	-0.62**	-0.13	-0.04	-0.09
12. Saltiness					1	0.32	0.48*	0.62**	0.19
13. Graininess						1	0.09	0.08	-0.11
14. Overall flavour intensity							1	0.56**	0.46*
15. Flavour desirability								1	0.44*
16. Foreign flavour intensity									1

*, **, *** = Significant at P<0.05, 0.01 and 0.001, respectively.

There were some significant correlations ($P<0.05$) within sensory attributes. Initial juiciness was highly and positively correlated with sustained juiciness of the product and negatively correlated with graininess (r = -0.57, $P>0.01$). Similarly sustained juiciness also negatively correlated with graininess (r = -0.62, $P>0.01$). Saltiness of starch-LFPB had played significant role on the scoring of overall flavour intensity and flavour desirability (r = 0.48, $P< 0.05$; r = 0.62, $P>0.01$). Another relationship was the correlation between foreign flavour and other flavour attributes. Foreign flavour intensity had significant and positive effect on the overall flavour intensity (r = 0.46, $P<0.05$) and the flavour desirability (r = 0.44, $P<0.05$).

4.2.3.8 Summary and conclusions

The primary objective of this research was to characterize the instrumental and sensory properties of low-fat pork bologna formulated with Kabuli and Desi starch and compare them with starch sources used in commercial practice (i.e. potato starch) and a starch that has a close relationship to the chickpea (i.e. pea starch). The addition of starch resulted in the reduction in expressible moisture, purges loss and increased the cooking yield which suggests the starch may have fully gelatinized and increased the functionality of the starch-meat system. In general Kabuli and Desi starch at 2.0% improved the water holding of the LFPB hence increasing the cooking yield, those values were similar to the effect of potato starch (2.0% level) and better than the pea starch (2.0%). Colour of the bologna was minimally affected by addition of starches in the LFPB at 1.0 to 2.0%. Generally, LFPB formulated with 2.0% Kabuli and Desi starch had higher instrumental TPA values than those with pea and potato starch. LFPB containing chickpea starch (either 1.0% or 2.0%) were given acceptable sensory texture and flavour scores similar to the control. Therefore, it appears feasible and effective to incorporate chickpea starch at current tested levels in low-fat emulsion type meat products for acceptable sensory merits with improved physicotextural properties.

5.0 SUMMARY AND CONCLUSIONS

The principal objective of this research was to examine the possible use of the Western Canadian chickpea in a low-fat meat product. To select one or two chickpea varieties well-suited for meat applications, the physico-chemical, functional and thermal properties of seed, flour, protein isolate and starch from six chickpea varieties (three Kabuli varieties: CDC Xena, CDC Frontier, and Amit, and three Desi varieties: CDC Cabri, CDC Vanguard, and Myles) were evaluated and compared to each other. To study the year effect, samples were collected from two production years (2005 and 2006).

Kabuli-type chickpea seed had higher seed weight, seed density, seed volume, hydration capacity and swelling capacity than did the Desi-type seed. However, the weight of the Desi seed coat was higher than that of the Kabuli-type. A strong positive correlation between seed weight and hydration capacity and swelling capacity was observed. Seed volume exhibited a highly significant positive correlation with swelling capacity and hydration capacity. A positive correlation between the water holding capacity and oil absorption capacity of chickpea flour and the seed weight, volume, hydration capacity and swelling capacity of chickpea seed was observed. Therefore, selected physico-chemical properties of chickpea seed might be useful in predicting the properties of particular varieties and of flours.

The chemical composition of chickpea flours showed that Kabuli-type flour had higher protein content, but lower amylose, insoluble dietary fibre and total dietary fibre contents than did Desi-type flour. Isolated chickpea protein contained 73-85% protein, and isolated starch was of high purity (93-98% starch). SDS-PAGE showed similar polypeptide/protein bands of 7S and 11S globulins for both chickpea flours and protein isolates. Chickpea flours pasted at

lower temperatures (62-68°C) than did chickpea starch (70-75°C). Both flours and starches from Desi-type chickpeas were characterized as having higher pasting temperatures (by ~2°C) than those from Kabuli-type samples. Nevertheless, the peak temperatures of the DCS thermograms of different chickpea flours were slightly higher (64-65°C) than those of the starches (59-61°C). An interesting finding was that both flours and starches from chickpea harvested in 2006 had higher DSC onset and peak temperatures (by ~ 4°C) than those from chickpea harvested in 2005. Protein in chickpea flours and protein isolates exhibited a high denaturation temperature (> 90°C), which would need to be achieved for protein gelation in food systems. However, the onset temperature of chickpea protein isolates, from small amplitude oscillatory testing, suggested that chickpea protein started to denaturate at a lower temperature (62-78°C). Study of the salt tolerance of chickpea protein isolates showed an optimum salt concentration of 1% for gel formation. Flours and protein isolates from different Kabuli cultivars had higher water holding capacities, oil absorption capacities, emulsion activities and stabilities than did corresponding products from Desi cultivars.

Overall, the results from flour, starch and protein characterization (study one) showed that chickpea had potential as a food ingredient with beneficial technological function. However, flours and fractions from CDC Xena (Kabuli-type) and Myles (Desi-type) had significantly better water and oil absorption, gelation capacity, and emulsification properties than did those from the other cultivars, hence they were chosen for further evaluation in a low fat meat system (bologna).

The results from study two showed that the incorporation of chickpea flour (2.5 and 5.0%) increased the yield and water holding properties of cooked low-fat pork bologna (LFPB). When the level of addition of flour was increased from 2.5 to 5.0%, product hardness also increased. For most sensory-flavour properties, products containing Kabuli or Desi chickpea flour performed similarly to the control. Bologna containing chickpea flour was juicier, firmer and had better flavour than did those containing wheat or pea flour (5.0%). Results from this

study indicated that chickpea flour at 2.5 and 5.0% (on wt basis) had potential as an extender in low fat meat systems.

Overall, low-fat pork bolognas containing 1.5 or 3.0% (on wt basis) chickpea protein did not exhibit altered textural and sensory characteristics. The addition of chickpea protein improved batter characteristics by increasing water holding properties and decreasing cooking loss. As the level of chickpea protein increased from 1.5 to 3.0%, bologna colour became more yellow and the texture became firmer. Chickpea protein at the 1.5% addition level did not alter the flavour properties of LFPB. These findings may allow increased usage of chickpea protein as a new plant ingredient in meat products.

Chickpea starch (2.0%) improved the water holding and cook yield of LFPB. Colour was minimally affected by starch addition. Generally, LFPB formulated with 2.0% chickpea starch had firmer texture than did LFPB containing pea or potato starch. Low-fat pork bologna containing 1-2% chickpea starch had acceptable sensory, texture and flavour scores, similar to those of the control. Chickpea starch could be incorporated in LFPB formulations without causing detrimental textural and sensory characteristics.

Finally, the chickpea ingredients (flour, starch and protein) generally improved the water holding and cook yield of LFPB. Furthermore, these products had texture and sensory properties similar, or even superior, to those of the controls.

6.0 FUTURE STUDIES

This study demonstrated the feasibility of using chickpea flour as a binder in LFPB. Further research is needed to identify the microbial stability of the LFPB in order to determine the shelf-life of the products. Chickpea flour used in this study contained seed hull. The properties of LFPB such as colour and sensory-graininess would be affected by seed coat residues. Therefore, an analysis of LFPB containing dehulled flour would be useful. To better understand water holding capacity, oil absorption capacity, cooking loss and many other textural properties of LFPB containing chickpea flour, protein or starch, investigation of gelation properties of these bolognas would be useful. In order to understand the mechanism of gelation of chickpea flour in meat systems, rheological studies, DSC work, scanning electron microscopic (SEM) and spectroscopic methods such as FTIR/IR and Raman studies would be helpful.

Since LFPB is a low-lipid emulsion system and chickpea protein is a good emulsifier, it would be beneficial to study emulsion activity and examine the factors affecting stability of high fat meat systems as well. Chickpea proteins had denaturation temperatures >90°C. Low-fat pork bolognas were cooked only to 72°C, hence chickpea proteins in this meat system would not be fully functional. In order to get full functionality of protein, one could prepare pretreated isolates which could gel at a lower temperature. Furthermore, the chickpea protein isolate that was used in this study had fat in it. It would be also interesting to know the effect of fat in chickpea protein isolates on the functionally of LFPB by formulating LFPB with defatted chickpea protein isolates. In order to understand the behavior of starch gelatinization in the meat matrix, further investigation needs to be done. According to most sensory attributes, sensory scores were not significantly affected by the starch levels used in this study. Hence, it would be desirable to study LFPB containing higher levels of starch (e.g. 2% or 4%).

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APPENDIX A:

DEFINITIONS AND TASTING PROCEDURE FOR THE SENSORY EVALUATION OF BOLOGNA

Evaluate the samples in the order that the scorecards are arranged. Circle a descriptor along the 8-point scale that best describes your impression of each of the characteristics.

Please take a drink of water before beginning and between samples. Unsalted crackers are also available as needed.

INITIAL JUICINESS- is the amount of fluid in the mouth during the first several chews

FIRMNESS- is the force required to bite through the sample with the incisors

COHESIVENESS- is the extent to which the sample was deformed between the teeth before it ruptures / breaks down

SUSTAINED JUICINESS- is the amount of moisture in the mouth after six to eight chews (towards the end of chewing).

SALTINESS- is the intensity of salty sensation after chewing

GRAININESS- is the presence of small particles after chewing

OVERALL FLAVOUR INTENSITY- is the amount of typical bologna seasoning and meat flavour present in the mouth after complete mastication

FLAVOUR DESIRABILITY- is the degree of liking of pleasantness of the flavor of the bologna

FOREIGN FLAVOUR INTENSITY- is the amount of any atypical or off-flavors present in the mouth after complete mastication (If any present, please describe in comments section)

COMMENTS: Your comments about each sample are welcome and would be very helpful. Before leaving the tasting session, please check to ensure that you have completed the entire scorecard.

Thank – You !

PANELIST _____ **SCORECARD FOR BOLOGNA - 2007** **SAMPLE No.** [ ] **P#** _____

DATE _____ **B#** _____

Evaluate the samples in the order that the scorecards are arranged. For each characteristic, circle a descriptor along the scales that best describes your impression. Feel free to provide any comments as well. Please take a drink of water before beginning and between samples. Unsalted crackers are also available as needed.

	8	7	6	5	4	3	2	1
TEXTURE: Initial Juiciness	Extremely juicy	Very juicy	Moderately juicy	Slightly juicy	Slightly dry	Moderately dry	Very dry	Extremely dry
Firmness	Extremely firm	Very firm	Moderately firm	Slightly firm	Slightly soft	Moderately soft	Very soft	Extremely soft
Cohesiveness	Extremely cohesive	Very cohesive	Moderately cohesive	Slightly cohesive	Slightly brittle	Moderately brittle	Very brittle	Extremely brittle
Overall Juiciness	Extremely juicy	Very juicy	Moderately juicy	Slightly juicy	Slightly dry	Moderately dry	Very dry	Extremely dry
	6	5	4	3	2	1		
Saltiness	Extremely salty	Very salty	Moderately salty	Slightly salty	Very slightly salty	Not detectable		
Graininess	Extremely grainy	Very grainy	Moderately grainy	Slightly grainy	Very slightly grainy	Not detectable		
	8	7	6	5	4	3	2	1
FLAVOR: Overall Flavour Intensity	Extremely intense	Very intense	Moderately intense	Slightly intense	Slightly bland	Moderately bland	Very bland	Extremely bland
Flavour Desirability	Extremely desirable	Very desirable	Moderately desirable	Slightly desirable	Slightly undesirable	Moderately undesirable	Very undesirable	Extremely undesirable
Foreign Flavour (describe below)	No foreign flavour	Very weak	Moderately weak	Slightly weak	Slightly intense	Moderately intense	Very intense	Extremely intense foreign flavour
OVERALL Acceptability	Extremely acceptable	Very acceptable	Moderately acceptable	Slightly acceptable	Slightly unacceptable	Moderately unacceptable	Very unacceptable	Extremely unacceptable

Comments